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RATES OF GROWTH OF IMMATURE DOUGLAS FIR AS SHOWN BY PERIODIC REMEASUREMENTS ON PERMANENT SAMPLE PLOTS[1]

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SCOPE AND METHODS OF STUDY

In the region west of the Cascade Range in Washington and Oregon immature even-aged forests of pure or almost pure Douglas fir (*Pseudotsuga taxifolia*) are of common occurrence. The total area of these immature or "second-growth" forests of Douglas fir, anywhere from 1 to 120 years of age, has been estimated at 4,500,000 acres at least. They stand partly on the national forests, but a good half of them are on private lands.

These forests originated naturally, either following severe fires which killed the former forest or, more rarely, after clear cutting in a logging operation. Some that followed forest fires are very extensive, even-aged, uniformly stocked with trees, and unbroken over thousands of acres; others are but small patches surrounded by timber of another age, or occasionally are a composite of several ages.

Rapid growing as they are, these stands are of tremendous economic importance in the region. Although not now merchantable for saw logs in the general market, they will furnish an acceptable supply of stumpage for the lumber industry when the virgin supplies of large timber are gone. They now afford the forester an unexcelled opportunity to study, by either the temporary-plot or permanent-plot method, the growth and yield of Douglas fir. Such studies are of value in determining not only how fast these particular forests are growing but also what yields may be generally expected after logging and natural reforestation. Many of the stands are so uniform and well stocked that they are a fair index of what may be expected under forest management.

A yield study of these stands by the temporary-plot method was begun by the Forest Service in 1909–1911, and was continued in a more comprehensive and complete fashion and for a wider range of sites and ages in 1924–25 by the Pacific Northwest Forest Experiment Station. The results of these field studies are now being compiled for publication by R. E. McArdle. The part of this paper regarding "normality" of stands is based upon the 1924–25 study. An attempt is made here to summarize the measurements up to date, to draw any conclusions which may be definite enough, and also to present several methods of analysis of permanent sample-plot material.

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From the beginning of these studies, permanently located sample plots were established to supplement the results obtained by the temporary plots. By 1927, 28 such permanent plots had been put in, and more are being established every year. Twelve of these plots have had two or more remeasureemnts.

The procedure followed in establishing the permanent sample plots was briefly as follows: Plots, preferably 1 acre in size, were laid out in typical well-stocked areas of even-aged second-growth Douglas fir. The boundaries were blazed, the corners marked by posts, and all the trees tagged by numbered metal tags at 4.5 feet above average ground level. The diameters at this point, breast height, were then measured by diameter tape or, in the earlier measurements, by calipers, and recorded by tree number. The heights of a number of trees were then taken, either by transit, Abney levels, or hypsometer, to form the basis for a height-on-diameter curve. These measurements, with additional descriptive notes, were recorded every five years, and a report on each of the groups of plots was prepared by those in charge of the project and placed on file with the Forest Service.

In order to make the results truly comparable, the whole series of measurements have been worked up under the same system. A few of the more important preliminary computations can be briefly outlined before the plot values are discussed. Anamorphic principles were used in fitting the height curves for successive periods. The most reliable set of heights was used as the basis for a graduating curve. In volume computation the diameters were grouped in single inch classes and the volumes taken from the new volume tables for second-growth Douglas fir of the Pacific Northwest, prepared for the yield study already mentioned. These tables give the total cubic foot contents inside bark of the tree; the International (⅛-inch kerf) board foot volume to a minimum d. b. h. (diameter at breast height) of 7 inches, a top diameter of 5 inches inside bark, and a 1.5-foot stump; and the Scribner board-foot volume to a minimum d. b. h. of 12 inches, top diameter inside bark of 8 inches, and stump height of 2 feet. No allowance for defect or waste in logging is made in any of the computations. All values are expressed in terms of the horizontal acre, although some of the plots were originally laid out by surface measure.

DESCRIPTION OF PERMANENT SAMPLE PLOTS

CASCADE PLOTS

In the spring of 1910 three 1-acre plots were located near Eula, Oreg., on the Cascade National Forest. (Fig. 1.) The stand was estimated to be 54 years old at that time. Recent investigation has determined the average condition of the land to be Site Quality II or, if the site-index system is used, site index 165. "Site index" is here defined as the height of the average dominant and codominant trees when the stand is 100 years of age. The basis for estimating the site index of plots younger or older than 100 years is found in curves drawn up for the Douglas fir yield study. Site-index values were computed for all periods, from which an average or most likely value was drawn. The site indices as computed showed an unexpected consistency for the several periods, any existent variability being chiefly attributable to the difference of estimators in judging crown classes.

The plots are located on a gentle north slope about 200 feet above the river valley. In this practically pure Douglas fir stand only a scattering of other species is to be found, such as madroño (*Arbutus menziesii*), golden chinquapin (*Castanopsis chrysophylla*), big-leaf maple (*Acer macrophyllum*), western hemlock (*Tsuga heterophylla*), and Pacific dogwood (*Cornus nuttallii*), all of which are in the understory. The stand is well closed and in 1910 had a crown density of

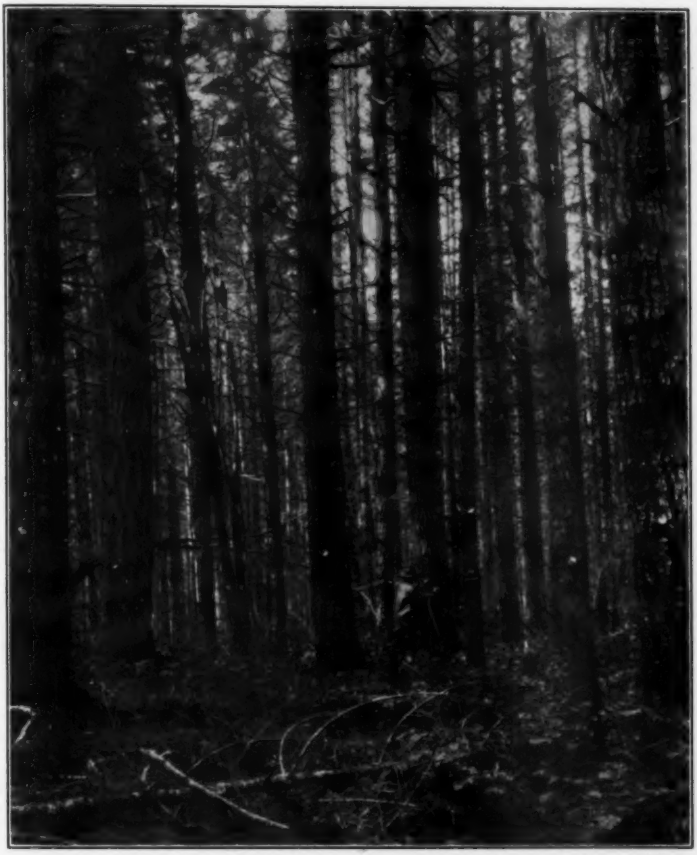

FIG. 1.—Cascade permanent sample plot No. 1, in a stand of Site Quality II, aged 69 years in 1925; volume 10,316 cubic feet, or 49,280 board feet by the Scribner rule

about 0.9. Several ice storms, notably one in 1888 and another in 1917, have produced bayonet tops and crooks and have killed some of the smaller trees. At the time of the establishment of the plots seedlings were quite numerous, but in the last measurement very few were found. Approximately three-quarters of the area is covered by underbrush, chiefly salal (*Gaultheria shallon*) and Oregon grape (*Berberis* sp.).

Since 1910 the plots have been remeasured at intervals of five years. In 1915 and 1920, however, the measurements were taken after the growth season was well advanced. For this reason the three periods instead of being taken as 5 growth years each, are assumed to be 5½, 5, and 4½ growth years, respectively. Table 1 lists the more important plot values per acre. Only live trees are included.

TABLE 1.—*Plot values per acre, permanent sample plots, Cascade National Forest* •

Year	Plot No.	Age	Number of conifer trees			Average d. b. h.	Average height	Total basal area		Volume of conifer trees		
			All	12 inches +	7 inches +			Conifers	Hardwoods	Cubic measure	International rule	Scribner rule
		Yrs.				*In.*	*Ft.*	*Sq. ft.*	*Sq. ft.*	*Cu. ft.*	*Bd. ft.*	*Bd. ft.*
1910	1	54	188	100	165	13.3	103	181.2	8.7	7,354	48,420	29,010
	2	54	214	124	193	13.5	104	212.5	1.6	8,706	58,770	34,710
	3	54	190	118	171	14.4	108	214.7	5.3	8,833	59,280	36,380
Average			197	114	176	13.7	105	202.8	5.2	8,298	55,490	33,367
1915	1	59½	175	106	159	14.3	116	194.2	4.0	8,709	60,270	37,440
	2	59½	198	127	189	14.6	117	231.7	.8	10,274	71,460	44,040
	3	59½	175	124	168	15.6	121	233.2	2.2	10,406	72,820	46,410
Average			183	119	172	14.8	118	219.7	2.3	9,796	68,183	42,630
1920	1	64½	149	110	143	16.0	126	207.0	3.5	9,528	68,590	44,070
	2	64½	160	122	160	16.1	126	226.9	.6	10,536	74,920	47,860
	3	64½	157	120	154	16.7	128	237.2	2.0	10,934	78,090	50,940
Average			155	117	152	16.3	127	223.7	2.0	10,333	73,867	47,623
1925	1	69	141	111	138	16.9	132	218.2	2.8	10,316	74,260	49,280
	2	69	148	123	148	17.2	132	238.7	.6	11,324	80,370	53,860
	3	69	147	120	147	17.7	134	251.3	1.7	11,864	86,070	57,290
Average			145	118	144	17.3	133	236.1	1.7	11,168	80,233	53,477

• Plots 1 and 2=1 acre, plot 3=0.98 acre; Site Quality II; site index 165, average for all periods.

In 1910 all diameters were measured with calipers. In 1915, the diameters on plots 1 and 2 were taken with calipers and on plot 3 with diameter tape; from then on the diameter tape was used exclusively. Since McArdle has found that diameter tape measurements overrun caliper measurements by approximately 1.5 per cent in basal area in stands of similar type and range of diameters, correction factors were applied to the values for the years when calipers were used. Basal areas and cubic foot volumes in Table 1 are increased by 1.5 per cent; International and Scribner board foot volumes by 2 per cent.

The periodic and mean annual increments are presented in Table 2.

TABLE 2.—*Annual increments per acre, permanent sample plots, Cascade National Forest*

PERIODIC ANNUAL INCREMENTS

Plot No.	Cubic measure			International rule			Scribner rule		
	First period	Second period	Third period	First period	Second period	Third period	First period	Second period	Third period
	Cu. ft.	*Cu. ft.*	*Cu. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*
1	246	164	175	2,155	1,664	1,260	1,533	1,326	1,158
2	285	52	175	2,307	692	1,211	1,696	764	1,333
3	286	106	207	2,462	1,054	1,773	1,824	906	1,411
Average	272	107	186	2,308	1,137	1,415	1,684	999	1,301

TABLE 2.—*Annual increments per acre, permanent sample plots, Cascade National Forest*—Continued

MEAN ANNUAL INCREMENTS

Plot No.	Cubic measure				International rule				Scribner rule			
	First measurement	Second measurement	Third measurement	Fourth measurement	First measurement	Second measurement	Third measurement	Fourth measurement	First measurement	Second measurement	Third measurement	Fourth measurement
	Cu. ft.	*Cu. ft.*	*Cu. ft.*	*Cu. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*
1	136	146	147	150	897	1,013	1,063	1,076	537	629	683	714
2	161	173	163	164	1,088	1,201	1,162	1,165	643	740	742	781
3	164	175	170	172	1,098	1,224	1,211	1,247	674	780	790	830
Average	154	165	160	162	1,028	1,146	1,145	1,163	618	716	738	775

If to the volume of the live trees the volumes of trees which died during any one period were added, a truer indication of total yield up to the end of the period would be obtained. Such volume is seldom considered in young natural, unthinned stands. At times, however, it does furnish a clue to unusual increments. Thus the apparently low increments for the period 1915–1920 are explained by the extraordinary losses by storms during that period. (Table 3.)

TABLE 3.—*Mortality by periods, permanent sample plots, Cascade National Forest*

Plot No.	Number of trees	Basal area	Volume		
			Cubic measure	International rule	Scribner rule
1911–1915		*Sq. ft.*	*Cu. ft.*	*Bd. ft.*	*Bd. ft.*
1	13	2.81	81	258	
2	16	4.36	131	545	
3	14	2.94	83	222	
Average	14.33	3.37	98	342	
Average per cent *		1.53	1.00	0.50	
1916–1920					
1	26	9.26	331	1,695	348
2	38	16.69	631	2,437	1,329
3	18	9.82	391	2,500	1,045
Average	27.33	11.92	451	2,211	907
Average per cent *		5.33	4.36	2.99	1.90
1921–1925					
1	8	2.70	96	505	0
2	12	7.88	328	2,159	777
3	10	4.02	150	815	140
Average	10	4.87	191	1,160	306
Average per cent *		2.06	1.71	1.45	0.57

* Percentages are expressed on the basis of plot values at the end of the period.

SIUSLAW PLOTS

Four plots on the Siuslaw National Forest have also been measured four times. In 1926 three new plots were laid out with the hope of supplementing the data for other sites and grades of stocking.

PLOTS 4 AND 5

In 1911 two plots were established in a 50-year-old Site Quality II stand (site indices 167 and 160) near Alpha, Oreg. Like most of the forests of this type, this one originated after a severe fire which

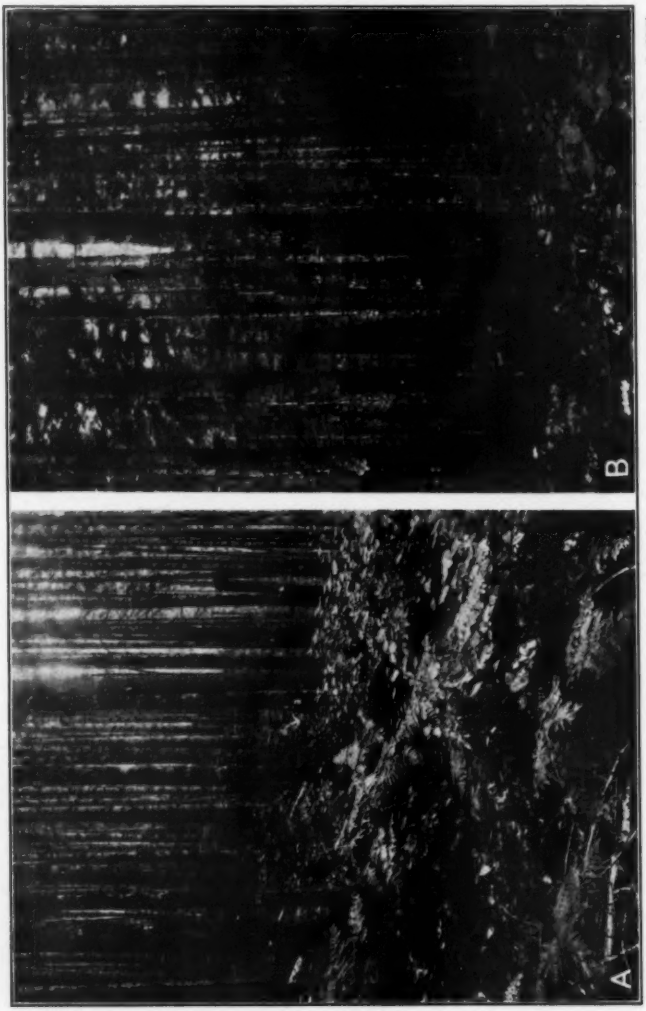

FIG. 2.—A, Permanent sample plot No. 5 on the Siuslaw National Forest, Site Quality II, aged 65 years in 1926; volume 12,397 cubic feet, or 47,800 board feet by the Scribner rule; many dead trees in stand. B, A companion plot to Siuslaw permanent sample plots Nos. 6 and 7; Site Quality I, aged 53 years in 1926, showing more even stocking than plots 6 and 7; present volume 10,964 cubic feet, or 44,400 board feet Scribner. Several stubs from the old fire which destroyed the original stand still remain

killed practically all of the former forest. Plot 4 is on a steep 60 per cent southwest slope and plot 5 (fig. 2, A) on a moderate 20 per sent south slope. There are no rock outcrops and the soil is a deep sandy loam. Stand density was originally given as 0.85–0.9. The underbrush and ground cover are now very abundant, salal, vine

maple (*Acer circinatum*), and ferns occupying much space. The stand is in good health, although signs of a previous ice storm are numerous.

The plots were remeasured in late summer or autumn at five-year intervals. Corrections for caliper measurements were made for the 1911 values.

Mortality on plot 5 was high in the second period, resulting in a low net cubic foot increment for this period. As the mortality was confined chiefly to the smaller diameters, board foot increment was not appreciably affected.

Plots 6 and 7

In the same year that plots 4 and 5 were laid out at Alpha, Oreg., two others were established in close proximity in a 38-year-old stand of Site I (site index 186) on Saddle Mountain, near Pawn, Oreg. Plot 6 is 1 acre and plot 7 is one-half acre in size. On each the slope and aspect are variable, since a small gully cuts through the plots. There are no rock outcrops and the soil is a deep sandy loam. The crown density of the stand was 0.75 at the time of establishment. Several holes contribute to an undesirable irregularity. The stand is in good health. A heavy underbrush and ground cover consisting chiefly of salal, together with sword fern, vine maple, and salmonberry (*Rubus spectabilis*), cover practically the whole area. Numerous old stubs, remnants of the original forest which was destroyed by fire, are found here and there.

Unfortunately the trees were not marked with metal tags until 1926. Pronounced systematic errors can arise where the trees are untagged, since the caliper man has no guide to where the diameter was last taken. A man might, for instance, take the diameters consistently low, thus increasing the diameters, the volumes, and the increments for that period. Such was evidently the case in 1921 on these two plots. Diameter, volume, and increment values are extremely erratic and must be omitted in almost all the discussions. In addition, no reliable record of mortality could be kept. In the tabulation, corrections for obtaining diameters by calipers instead of by diameter tape are made for the years 1911 and 1916. Plot values per acre and annual increments are shown in Tables 4 and 5; mortality records for plots 4 and 5 are shown in Table 6.

TABLE 4.—*Plot values per acre, permanent sample plots, Siuslaw National Forest* [a]

Year	Plot No.	Age	Number of conifer trees			Average d. b. h.	Average height	Total basal area		Volume of conifer trees		
			All	12 inches +	7 inches +			Conifers	Hardwoods	Cubic measure	International rule	Scribner rule
		Yrs.				*In.*	*Ft.*	*Sq. ft.*	*Sq. ft.*	*Cu. ft.*	*Bd. ft.*	*Bd. ft.*
1911	4	50	298	185	273	13.4	102	292.8	11.9	11,980	79,290	48,350
	5	50	382	110	336	10.65	102	236.3	0	9,896	63,390	26,370
Average			340	148	304	11.95	102	264.6	6.0	10,938	71,340	37,360
	6	38	199	101	181	13.1	95	187.2	3.8	7,061	44,920	25,280
	7	38	236	82	202	10.8	87	151.0	5.7	5,446	32,520	14,440
Average			218	92	192	11.9	91	169.1	4.8	6,254	38,720	19,860
1916	4	55	273	192	263	14.9	112	329.4	9.1	14,152	96,870	61,620
	5	55	347	134	331	11.6	109	255.1	0	11,180	74,450	35,500
Average			310	163	297	13.15	110	292.2	4.6	12,666	85,660	48,560
	6	43	183	115	177	14.45	105	208.2	3.6	8,417	56,130	33,760
	7	43	161	93	161	13.3	102	156.2	9.2	6,272	40,920	22,120
Average			172	104	169	13.95	104	182.2	6.4	7,344	48,525	27,940
1921	4	60	238	190	233	16.35	120	346.6	6.7	15,520	109,000	71,870
	5	60	297	145	290	12.6	116	256.3	0	11,626	79,590	42,290
Average			268	168	262	14.4	118	301.4	3.4	13,573	94,295	57,080
	6	48	156	131	153	17.0	119	245.6	0	10,685	74,720	49,620
	7	48	138	120	116	16.0	116	192.8	0	8,381	58,120	37,620
Average			147	126	134	16.5	118	219.2	0	9,533	66,420	43,620
1926	4	65	228	192	223	17.1	125	364.1	6.5	16,888	120,450	81,510
	5	65	286	152	279	13.1	119	267.0	0	12,397	86,210	47,890
Average			257	172	251	15.0	122	315.6	3.2	14,642	103,330	64,700
	6	53	150	131	149	17.2	127	241.9	5.9	11,223	80,110	53,880
	7	53	132	104	132	16.3	124	190.4	4.5	8,806	62,320	39,880
Average			141	118	140	16.8	126	216.2	5.2	10,014	71,215	46,880

[a] Plot 4=0.399 acre, plot 5=0.455 acre, plot 6=0.932 acre, plot 7=0.441 acre. Plots 4 and 5 are Site Quality II (site indices 167 and 160), plots 6 and 7 are Site Quality I (site index 186)

TABLE 5.—*Annual increments per acre, permanent sample plots, Siuslaw National Forest*

PERIODIC ANNUAL INCREMENTS

Plot No.	Cubic measure			International rule			Scribner rule		
	First period	Second period	Third period	First period	Second period	Third period	First period	Second period	Third period
	Cu. ft.	*Cu. ft.*	*Cu. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*
4	434	274	274	3,516	2,426	2,290	2,654	2,050	1,928
5	257	89	154	2,212	1,028	1,324	1,826	1,358	1,120
Average	346	182	214	2,864	1,727	1,807	2,240	1,704	1,524
6	271	454	108	2,242	3,718	1,078	1,696	3,172	852
7	165	422	85	1,680	3,440	840	1,536	3,100	452
Average	218	438	96	1,961	3,579	959	1,616	3,136	652

TABLE 5.—*Annual increments per acre, permanent sample plots, Siuslaw National Forest*—Continued

MEAN ANNUAL INCREMENTS

Plot No.	Cubic measure				International rule				Scribner rule			
	First measurement	Second measurement	Third measurement	Fourth measurement	First measurement	Second measurement	Third measurement	Fourth measurement	First measurement	Second measurement	Third measurement	Fourth measurement
	Cu. ft.	*Cu. ft.*	*Cu. ft.*	*Cu. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*
4	240	257	259	260	1,586	1,761	1,817	1,853	967	1,120	1,198	1,254
5	198	203	194	191	1,268	1,354	1,326	1,326	527	645	705	737
Average	219	230	226	225	1,427	1,557	1,572	1,590	747	883	951	995
6	186	196	223	212	1,182	1,305	1,557	1,512	665	785	1,034	1,017
7	143	146	175	166	856	952	1,211	1,176	380	514	784	752
Average	165	171	199	189	1,019	1,128	1,384	1,344	523	650	909	885

TABLE 6.—*Mortality, by periods, permanent sample plots, Siuslaw National Forest*

1912–1916

Plot No.	Number of trees	Basal area	Volume		
			Cubic measure	International rule	Scribner rule
		Sq. ft.	*Cu. ft.*	*Bd. ft.*	*Bd. ft.*
4	25	7.67	241	1,075	
5	37	7.67	169	563	
Average	31	7.67	205	819	
Average per cent [a]		2.62	1.62	0.96	

1917–1921

4	35	12.63	418	2,195	
5	51	16.70	576	3,084	
Average	43	14.66	497	2,640	
Average per cent [a]		4.86	3.66	2.80	

1922–1926

4	10	5.54	214	1,145	501
5	11	4.29	163	936	
Average	10.5	4.92	188	1,040	250
Average per cent [a]		1.56	1.28	1.01	.39

[a] Percentages are expressed on the basis of plot values at the end of the period.

COLUMBIA PLOTS

Five plots were laid out in 1914 on the Columbia National Forest in several localities and sites, and were measured successively in 1914, 1919, and 1924. Two of the plots had to be abandoned because a road was built through them in 1923. In 1924 one new plot was put in to take their place. The 1924 measurements were taken in the fall; the two previous ones were taken in the spring. The last period, therefore, contains six growing seasons.

Plots 1, 2, and 3

Along the old Race Track Trail from Hemlock Ranger Station in the Wind River Valley, Wash., three 1-acre plots were laid out in a fairly thrifty 72-year-old stand, which came in after a fire made a clean burn of the original forest. Conditions are variable and consequently the site is not the same for each plot. Plot 1 is in a Site IV stand (site index 110), and plots 2 and 3 are in Site III stands (site indices 150 and 133). Plots 1 and 2 are on gentle slopes, while plot 3 is on a steep 60 per cent slope. The soil is a deep sandy loam with some gravel and small stones. The crown density varied from 0.6 to 0.8 at the time of establishment, and the trees are uniformly distributed over the plots. The underbrush and ground cover are light and consist of scattered dogwood, western mountain ash (*Sorbus sitchensis*), vine maple, Oregon grape, fern, goatbrush (*Pachistima myrsinites*), and other species in lesser quantity.

All the plots were measured by calipers in 1914 and 1919, and plot 2, the only one remaining, by diameter tape in 1924. Corrections were applied for 1914 and 1919 to the existent plot 2 so as to bring all measurements on a comparable basis. Compared to that on some of the other plots, the death rate is rather low.

Plots 4 and 5

In a forest of similar age but on another drainage a few miles away, two more plots were established in 1914 in a stand on Site III (site index 144). Plot 4 is on a gentle to moderate west slope and plot 5 on a steep 65 per cent east slope. The soil is a loam underlain, on plot 5 at least, by bowlders. Underbrush and ground cover consist chiefly of vine maple, dogwood, Oregon grape, and fern in varying densities, but never very heavy. The crown density at the time of establishment was 0.7–0.8 on plot 4 and 0.6 on plot 5. The trees are thrifty and are fairly evenly distributed. One or two small holes now present in plot 4 were caused by the death of a few trees. The mortality in this plot has been higher than in any of the other Columbia plots.

Records for the five plots are presented in Tables 7 to 9.

Table 7.—*Plot values per acre, permanent sample plots, Columbia National Forest*[a]

Year	Plot No.	Age	Number of conifer trees			Average d. b. h.	Average height	Total basal area of conifers	Volume of conifer trees		
			All	12 inches +	7 inches +				Cubic measure	International rule	Scribner rule
		Yrs.				*Inches*	*Feet*	*Sq. ft.*	*Cu. ft.*	*Bd. ft.*	*Bd. ft.*
1914	1	72	251	120	224	12.1	85	200.8	6,904	40,910	21,660
	2	72	160	116	146	16.0	113	223.0	9,413	64,660	43,060
	3	72	246	146	217	13.5	99	244.1	9,393	61,910	37,920
	4	72	185	127	177	14.7	110	218.1	9,089	61,130	38,160
	5	72	157	127	150	16.6	112	236.0	9,852	67,820	44,980
1919	1	77	243	130	220	12.8	94	214.9	7,888	48,930	26,770
	2	77	149	117	141	17.2	124	239.8	10,786	76,820	51,410
	3	77	241	159	213	14.5	109	276.3	11,354	76,350	48,500
	4	77	176	133	170	15.6	113	234.6	10,201	71,090	45,570
	5	77	154	130	147	17.5	122	257.3	11,425	81,120	55,020
1924	1										
	2	83	145	121	139	18.0	131	256.1	11,953	86,290	58,850
	3										
	4	83	171	133	166	16.4	126	249.8	11,348	80,510	52,440
	5	83	151	132	148	18.3	130	276.2	12,862	92,900	64,050

[a] Plot 1=1 acre; plot 2=0.995 acre; plot 3=0.936 acre; plot 4=0.975 acre; plot 5=0.938 acre. Plot 1=Site Quality IV (site index 110); plots 2 to 5=Site Quality III (site indices 150, 133, 144, and 144, respectively).

TABLE 8.—*Annual increments per acre, permanent sample plots, Columbia National Forest*

PERIODIC ANNUAL INCREMENTS

Plot No.	Cubic measure		International rule		Scribner rule	
	First period	Second period	First period	Second period	First period	Second period
	Cu. ft.	*Cu. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*
1	197		1,604		1,022	
2	275	194	2,432	1,578	1,670	1,240
3	392		2,888		2,116	
4	222	191	1,992	1,570	1,482	1,145
5	315	240	2,660	1,963	2,008	1,505

MEAN ANNUAL INCREMENTS

Plot No.	Cubic measure			International rule			Scribner rule		
	First measurement	Second measurement	Third measurement	First measurement	Second measurement	Third measurement	First measurement	Second measurement	Third measurement
	Cu. ft.	*Cu. ft.*	*Cu. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*	*Bd. ft.*
1	96	102		568	635		301	348	
2	131	140	144	898	998	1,040	598	668	709
3	130	147		860	992		527	630	
4	126	132	137	849	923	970	530	592	632
5	137	148	155	942	1,054	1,119	625	715	722

TABLE 9.—*Mortality by periods, permanent sample plots, Columbia National Forest*

1915–1919

Plot No.	Number of trees	Basal area	Volume		
			Cubic measure	International rule	Scribner rule
		Sq. ft.	*Cu. ft.*	*Bd. ft.*	*Bd. ft.*
1	8	1.32	30	73	
2	11	2.80	73	283	
3	4	1.62	46	226	
4	8	4.78	177	1,075	500
5	3	1.80	57	320	107
Average per cent [a]		1.01	0.75	0.56	0.27

1920–1924

2	4	1.86	59	302	
4	5	2.62	100	622	303
5	2	1.17	38	212	
Average per cent [a]		0.72	0.54	0.44	0.17

[a] Percentages are expressed on the basis of plot values at the end of the period.

DISCUSSION OF PLOT VALUES

VOLUMES

The total yields up to the time of last measurement, an important point of comparison, if arranged by site classes and rounded off to the nearest thousand can be quickly summarized. The Site I plots (Siuslaw 6 and 7), present age 53 years, have average volumes for

cubic foot, international board foot, and Scribner board foot of 10,000, 71,000, and 47,000, respectively. Three of the Site II plots (Cascade 1, 2, 3), age at last measurement 69 years, have average values of 11,000, 80,000 and 53,000 for these three standards of measurement, while the other two Site II plots (Siuslaw 4 and 5), age 65 years, carry 15,000, 103,000, and 65,000 feet. Disproportionately large values are caused by the inclusion of plot 4. All the way through, this plot has appeared erratically high. This is due partly to the fact that the plot was laid out by surface measure, and the steep slope upon which it lies necessitates a large area correction, increasing thereby all values on the plot.

Furthermore, the upper edge of the plot is along a ridge upon which is a strip of trees of large diameter. It takes only a few of such diameters to increase the volumes tremendously. If plot 5, which is more uniform than plot 4, is considered by itself the agreement with the Cascade plots is pronounced. The Site III plots which still exist (Columbia 2, 4, 5), age 83 years at last measurement, have an average cubic foot volume of approximately 12,000 feet, International board foot volume of 87,000 feet, and Scribner board foot volume of 58,000 feet. The relation of these values to the volumes as determined in the yield study for second-growth Douglas fir will be discussed later.

INCREMENTS

An inspection of the various periodic increment tables serves to show how very unstable such values are. Any unusual amount of mortality is reflected immediately, especially in the cubic foot volume increments which are so largely made up from the smaller trees having no appreciable board-foot volumes but significant cubic foot volumes. Naturally the mean annual increments run much more regularly. The mean annual increments of the Site III plots (Columbia) with the present age of 83 years are still increasing in all volumes; those of the Site II plots (Cascade 1, 2, and 3, Siuslaw 4 and 5, omitting 6 and 7), present ages 69 and 65 years, respectively, have practically if not certainly reached their maximum in cubic foot and International board-foot volume increments but are still increasing significantly in Scribner volume increments; those of the young Site I stands (now 53 years old) have not reached their peak as yet. However, for all the plots, with the exception of the doubtful plots (Siuslaw 6 and 7), the periodic annual increments are on the whole decreasing. Mortality during the middle period on the Cascade plots disguises the effect, but does not leave much doubt, especially if the diameter growth of the individual diameter classes is observed.

BOARD FOOT-CUBIC FOOT RATIOS

Within certain limits board foot-cubic foot ratios increase steadily with age, as is apparent in the tabulation of ratios. (Table 10.) The International board foot-cubic foot ratios are still increasing slowly, but have reached a position where further increase will be very small, especially in the older plots. Scribner ratios, however, continue to enlarge at a somewhat faster rate, and undoubtedly will continue to do so until the 12-inch diameter has been reached by all trees, this diameter being the minimum for which Scribner volumes were computed, as compared to 7 inches for International volumes.

TABLE 10.—*Board foot–cubic foot ratios*

Plot No.	International rule				Scribner rule			
	First measurement	Second measurement	Third measurement	Fourth measurement	First measurement	Second measurement	Third measurement	Fourth measurement
Cascade:								
1	6.58	6.92	7.20	7.20	3.94	4.30	4.63	4.78
2	6.75	6.96	7.11	7.10	3.99	4.29	4.54	4.76
3	6.71	7.00	7.14	7.25	4.12	4.46	4.66	4.83
Columbia:								
1	5.92	6.20	----	----	3.14	3.39	----	----
2	6.87	7.12	7.22	----	4.57	4.77	4.92	----
3	6.59	6.72	----	----	4.04	4.27	----	----
4	6.73	6.97	7.09	----	4.20	4.47	4.62	----
5	6.88	7.10	7.22	----	4.56	4.82	4.98	----
Siuslaw:								
4	6.62	6.84	7.02	7.13	4.04	4.35	4.63	4.83
5	6.40	6.66	6.85	6.95	2.66	3.18	3.64	3.86
6	6.36	6.67	7.69	7.14	3.58	4.01	4.64	4.80
7	5.97	6.52	6.93	7.08	2.65	3.53	4.49	4.53

RELATION OF PERMANENT SAMPLE-PLOT DATA TO YIELD TABLES

The degree of normality of these plots or the relation of the plot values to values given in the normal yield tables for second-growth Douglas fir leads to a number of conclusions which may prove valuable in the application of such tables, or at least suggest the nature of the results which can be obtained from a more detailed study. (Table 11.)

TABLE 11.—*Relation of permanent sample-plot data to normal yield tables*

[Values from sample-plot data are shown in percentage of normal yield for the four successive measurements]

Plot No.	Number of trees				Basal area				Volume by International rule				Volume by cubic measure				Volume by Scribner rule			
	First measurement	Second measurement	Third measurement	Fourth measurement	First measurement	Second measurement	Third measurement	Fourth measurement	First measurement	Second measurement	Third measurement	Fourth measurement	First measurement	Second measurement	Third measurement	Fourth measurement	First measurement	Second measurement	Third measurement	Fourth measurement
Cascade:																				
1	66	72	72	70	80	83	84	85	81	89	88	88	78	83	84	84	94	97	97	94
2	76	81	73	73	94	98	92	93	98	115	96	96	92	98	92	93	113	114	105	103
3	68	72	72	72	96	98	96	98	99	108	101	102	94	99	97	98	118	120	112	109
Columbia:																				
1	64	68	----	----	94	98	----	----	106	114	----	----	98	106	----	----	145	144	----	----
2	68	70	74	----	88	92	97	----	84	93	96	----	83	90	94	----	96	100	101	----
3	86	92	----	----	102	111	----	----	100	114	----	----	97	110	----	----	118	131	----	----
4	72	77	80	----	88	91	94	----	85	94	96	----	84	90	94	----	94	100	100	----
5	63	68	71	----	94	100	104	----	94	105	110	----	92	100	106	----	112	120	122	----
Siuslaw:																				
4	98	102	101	105	137	140	144	146	149	156	154	154	130	146	148	146	188	184	180	171
5	113	118	113	120	110	112	108	108	127	128	120	116	119	120	114	112	122	122	118	111
6	62	66	66	70	100	103	135	110	122	117	128	115	100	107	118	109	169	144	158	130
7	73	60	59	62	81	77	89	83	88	85	100	90	76	80	92	86	96	94	116	96

In the yield study for Douglas fir, "normality" is not defined as the maximum volume that may be produced by intensive care and treatment of stands, but as the volume produced under conditions of average full stocking of stands which remain unthinned throughout their lives. The conception of "normality" of the stand and the most appropriate index of "normality" raise questions which have

been debated frequently. Some identifying character or index is needed whereby the normality of the stand may be easily recognized. Several tests have been made to determine the best index of normality; such an index as might be employed when yield tables are to be applied to stands or portions of stands which are other than 100 per cent stocked or in full conformity to yield-table values. For this purpose total number of trees per acre has at times been used with other species in other regions.

Douglas fir percentages of normal number of trees did not conform at all to the percentages of normal volumes as indicated by the permanent sample plots. On the other hand, the relation of percentages of normal basal area and the percentages of normal volumes (see fig. 3) took trends similar to those obtained with number of trees in a yield study which the writer conducted in red spruce stands of the northeastern United States.

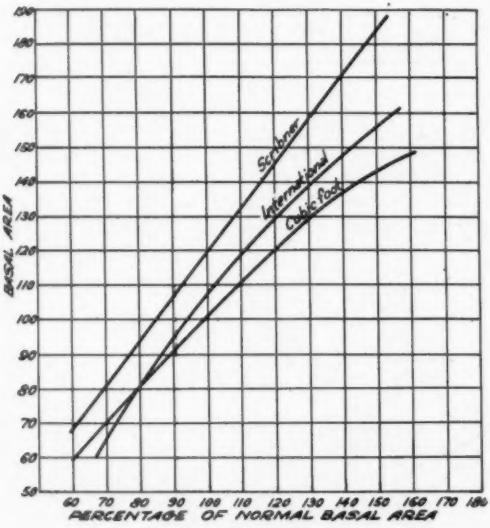

FIG. 3.—Relation of plots to normal yield tables

If a graph similar to Figure 3 be drawn with number of trees as index of normality, and using values in Table 11, several interesting points are brought out. An irregular distribution of points results in a large scatter for the lower percentages (number of trees) and abnormal relations in the upper percentages. If the plots conformed strictly to the yield tables, without considering the average or standard deviations of the table values, one would expect that when number of trees is 100 per cent the curves would pass through 100 per cent basal area and 100 per cent volume. Such is not the case, for, at 100 per cent number of trees, the percentages, especially those for the Scribner volume, run considerably higher. The extreme scatter of the points does not permit the placing of a satisfactory curve.

Indirectly this lack of coordination in normality percentages is indicative of variability in diameter distribution and spacing of trees in a plot, which the most careful selection of plots seldom overcomes.

If now the trend of volume percentages on basal area percentages is studied, an entirely different situation obtains. (Fig. 3.) The relation between cubic foot percentage and basal area percentage is extremely regular. Between International board-foot volume and basal area it is less regular but is still well defined. The 100–100 per cent is closely approached by the curved cubic-foot volume percentages, while International volume passes within a +9 per cent. Both in-

crease regularly in roughly parallel trends with the increase in basal area. The standard errors of the curves are ±4.53 per cent for the cubic foot-basal area relation and ±6.18 per cent for the International-basal area relation. Scribner-basal area relation is too weak (SE = ±13.66 per cent) to be of much value.

The curves in Figure 3 are based not only on data drawn from the 12 plots which have had at least two measurements, but also on data of all plots established previous to 1927, totaling 20 in number.

So far the relationships have been treated only as static measurements. It remains to determine whether they are static throughout the life of the stands or whether there is a fast or slow advance to normality of stocking. A simple way to observe this trend is to divide the stands into groups of normality percentages and to obtain average percentage rise or fall in stocking with each five-year period. The measurements of Siuslaw plots 6 and 7 and of the second period of Cascade plot 2 are omitted, leaving 22 periods as a working basis. This procedure is justifiable since some of the values are known to be erratic and not representative of normal growth.

From Table 12 approach towards normality is evident. Below 100 per cent stocking in any of the factors the trend is up to 100 per cent, but above 100 per cent stocking it is irregular, sometimes up and sometimes down. For instance, if an acre is below 100 per cent in basal area, in five years it will stand 2.9 per cent higher in the scale of normality; if deficient in International board-foot volume, the increase in normality percentage in five years will be 6.1 per cent. An increased number of period measurements is necessary before such values can be more certainly defined and curvilinear relations assigned.

TABLE 12.—*Approach to normality expressed in percentage increase for each five-year period*

Percentage of normality at beginning of period	Number of trees		Cubic measure		Basal area		Volume			
							International rule		Scribner rule	
	Number periods	Percentage rise or fall	Number periods	Percentage rise or fall	Number periods	Percentage rise or fall	Number periods	Percentage rise or fall	Number periods	Percentage rise or fall
	1	2	1	2	1	2	1	2	1	2
60–69	6	+4.0								
70–79	9	+1.7	1	+5.0						
80–89	1	+6.0	4	+3.5	5	+2.4	5	+5.0		
90–99	1	+4.0	10	+4.8	9	+2.8	6	+7.0	5	+2.0
100–109	2	+1.5	1	+6.0	3	+4.3	5	+4.2	3	−0.3
110–119	3	+2.3	2	−0.5	2	−1.0			6	+2.3
120–129			1	−6.0			3	−3.7	4	−2.5
130–139			1	+16.0	1	+3.0				
140–149			2	0.0	2	+3.0	1	+7.0	1	−1.0
150–159							2	−1.0		
180–189									3	−5.6
	22		22		22		22		22	
Average below 100 per cent		+2.9		+4.5		+2.7		+6.1		+2.0
Average above 100 per cent		+2.0		+2.1		+2.5		+1.4		−0.9
Total average		+2.7		+3.7		+2.6		+3.7		−0.2

This change in position relative to the normal yield tables affects directly the increments, especially the periodic annual increments. A larger periodic increment can be expected on the understocked plots than is indicated by the yield tables. For instance, the ages and the site represented in the Cascade plots should have approximately periodic annual cubic foot volume increments ranging between 210 and 170 cubic feet, decreasing for 15 years, while actually they average 185 cubic feet per year for the 15 years in spite of the great loss during the middle five-year period.

AVERAGE DIAMETERS AND DIAMETER GROWTH

The average diameters of the plot groups indicate rapid growth. The Site I Siuslaw plots 6 and 7 increased from 11.9 to 16.8 inches, an average of approximately 1 inch every 3 years. The Site II Cas-

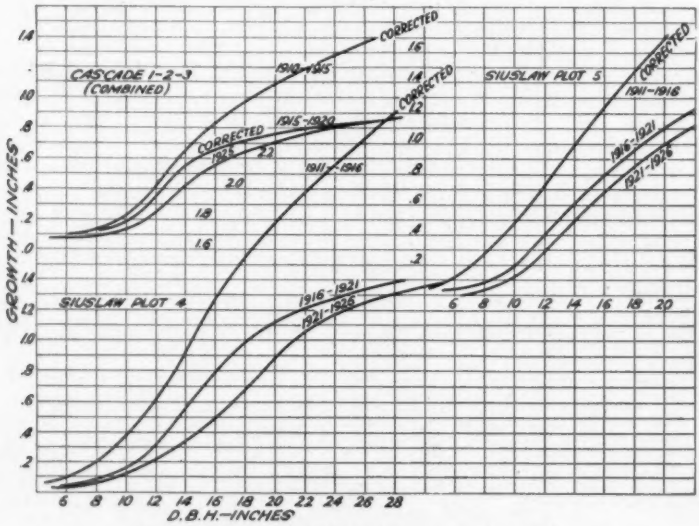

FIG. 4.—Diameter growth by inch classes, five-year periods

cade plots as an average increased from 13.7 inches to 17.3 inches in 15 years, which amounts to more than 1 inch every 5 years. The Site II Siuslaw 4 and 5 plots increased from 11.95 to 15 inches, or approximately 1 inch every 5 years. Site III Columbia plots 4 and 5 grew from 15.6 to 17.4 and plot 2 from 16 to 18 inches in 11 years, or an average of 1 inch every 6 years.

The rate of change of the average diameter of the plot is, however, no index of the rate of change of the diameters of the individual trees. From the original tallies of a number of the plots, the actual diameter growth was recorded and averaged for each period by diameter classes. (Fig. 4.) The diameter increment in all the diameter classes is steadily decreasing. This can usually be expected in stands which are left untreated by artificial thinning and subjected only to natural thinning.

Siuslaw plot 4 has been used in this series of graphs to illustrate definitely how the inclusion of exceptional diameter growth increases the periodic annual increments of the first period. An allowance for caliper readings which was made before the values were plotted has not resulted in making a much more normal diameter relation for the first period. Siuslaw plot 5 effects a similar but less pronounced distortion.

DISTRIBUTION OF TREES IN DIAMETER CLASSES

The distribution of diameters in a stand is thought to follow certain definite laws. Whether this distribution is in strict accordance with the law of probabilities or whether there exists some tendency to asymmetry or skewness is a question to be solved for each

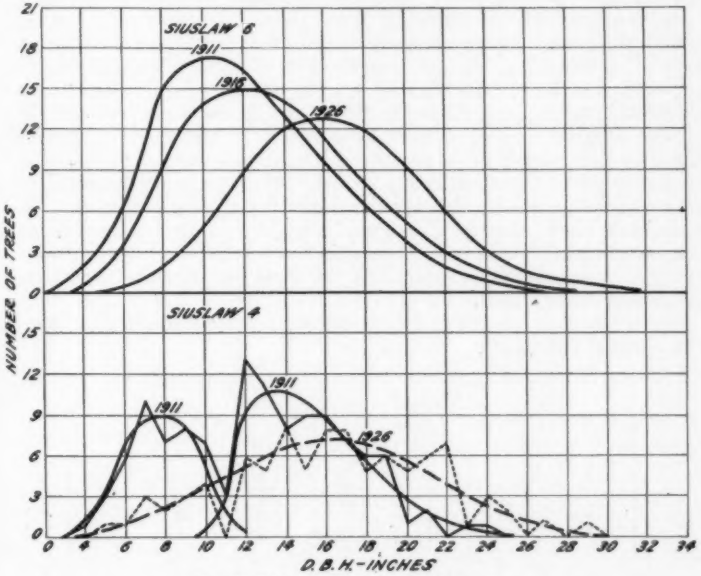

FIG. 5.—Distribution of diameters in stands

individual species. It seems natural to presume that the more tolerant a species is, the longer will the smaller diameters hold on to life and the more skewed to these diameters will the distribution be. Also the younger a plot is, the more will the distribution of trees in diameter classes be skewed to the smaller diameters. Such a situation can be shown graphically, as in the frequency curve (fig. 5) of Siuslaw plot 6 at ages 38, 43, and 53. Age 48 has been left out as being undeniably erratic. The curves shown in the graph were obtained by drawing a smooth curve through a histogram illustrating the number of trees in each diameter class plotted upon the diameter as abscissa. They fit the distribution as closely as can be judged ocularly. The number of trees is the actual number on the plot and not the number per acre. It is obvious even from such a rough graph that there is a change in the character of diameter distribution with the

age of the stand. Similar effects are obtained on all plots. The first and last stages of diameter distribution shown for Siuslaw plot 4 indicate a unique case. Apparently there are two distinct frequencies at the beginning which gradually lose their distinction with age and are finally combined into one fairly normal curve. This double frequency is not due to difference in age but to the very apparent difference of spacing, which affects the diameter development within the plot, as previously mentioned. If Columbia 2 were plotted, an asymmetry of opposite nature to Siuslaw 4 would be found. The question now arises how such distributions can be more accurately defined.

The following method has been adapted from Charlier's method of computation for skewed frequencies made use of by Y. Ilvessalo in a Finnish publication.[2] Of all theoretical fittings of distribution, it seems about the most easily applicable. In essence, the method consists in computing from each distribution two coefficients (coefficient of asymmetry, B_3, and coefficient of excess, B_4) which, when applied to a definite formula and to a set of tables known as Charlier's tables, will result in giving a theoretically fitted distribution. With a large number of plots, the plots could be grouped by desirable classes and a stand table made which would then be subjected to the first part of the treatment, namely, through the computation of the coefficients up to the application of Charlier's tables. These coefficients could then be averaged or plotted in any desired manner, such as plotting on age, or averaging by type, and the curved or averaged coefficient values could be used for obtaining a correct theoretical stand table.

The scheme of computation is shown applied to Siuslaw plot 5, 1916 (area 0.455 acre).

D. b. h.	Class (c)	Number of trees (NT)	c(NT)	c^2(NT)	c^3(NT)	c^4(NT)
5	−6	1	−6	36	−216	1,296
6	−5	6	−30	150	−750	3,750
7	−4	16	−64	256	−1,024	4,096
8	−3	24	−72	216	−648	1,944
9	−2	19	−38	76	−152	304
10	−1	16	−16	16	−16	16
11	0	15	0	0	0	0
12	+1	13	13	13	13	13
13	2	9	18	36	72	144
14	3	8	24	72	216	648
15	4	11	44	176	704	2,816
16	5	7	35	175	875	4,375
17	6	3	18	108	648	3,888
18	7	5	35	245	1,715	12,005
19	8	2	16	128	1,024	8,192
20	9	1	9	81	729	6,561
21	10	1	10	100	1,000	10,000
22	11	1	11	121	1,331	14,641
Sums=S		158	−226	+2,005	−2,806	+74,689
			+233		+8,327	
			+7		+5,521	

The first step in the computation is to list the number of trees by diameter classes, then to give to the diameter group that is estimated

[2] ILVESSALO, Y. TUTKIMUKSIA METSÄTYYPPIEN TAKSATOORISESTA MERKITYKSESTÄ. [INVESTIGATIONS ON THE IMPORTANCE OF FOREST TYPES IN FOREST MENSURATION AND VALUATION.] Acta Forest. Fennica 15, 157 p., illus. 1920. (Separately paged.) [German résumé, 26 p.]

to be near the average value the class value of zero and to all other diameter classes progressive plus or minus values, as shown in the illustration. The fourth column is obtained by multiplying the class value by the number of trees in that class; the fifth column is obtained by multiplying the square of the class value by the number of trees in the class, and so on for the remaining columns. The columns are summed up with strict consideration of the plus and minus signs. Following this, a second set of computations is made.

$$d_1 = \frac{\text{Sum } c(NT)}{\text{Sum } (NT)} = \frac{+7}{158} = .044$$

$$d_2 = \frac{\text{Sum } c^2(NT)}{\text{Sum } (NT)} = +12.690$$

$$d_3 = \frac{\text{Sum } c^3(NT)}{\text{Sum } (NT)} = 34.943$$

$$d_4 = \frac{\text{Sum } c^4(NT)}{\text{Sum } (NT)} = 472.715$$

$$d_1^2 = +.002$$

$$d_1^3 = +.000$$

$$d_1^4 = +.000$$

$$\text{Average DBH} = 11 + d_1 = 11.044$$

Standard deviation squared =

$$SD_1^2 = d_2 - d_1^2 = 12.690 - .002 = 12.688$$

$$SD_1 = 3.562$$

$$SD_1^3 = 45.195$$

$$SD_1^4 = 160.985$$

Total basal area of plot (square feet)

$$= (NT) \times \frac{.7854 \text{ (av. DBH}^2 + SD_1^2)}{144}$$

$$= 158 \times \frac{.7854 \text{ (134.658)}}{144} = 116.04 \text{ square feet}$$

The above shows first the computation of several deviation values (d_1, d_2, d_3, d_4), arrived at by dividing the fourth, fifth, sixth, and seventh columns of the first tabulation by the total number of trees. The actual average diameter of the stand is found by adding to the assumed average diameter the value of d_1, taking the sign into consideration. The square of the standard deviation (SD) is obtained by subtracting from d_2 the square of d_1. The total basal area of the stand is found by multiplying the area of a circle the square of whose diameter is equal to the square of the average diameter of the stand, plus the square of the standard deviation, by the total number of trees. This method of computing total basal area checks out very

closely with the value obtained in the ordinary way. A further calculation must now be made.

		Check
$d_3 = +34.943$	$d_4 = 472.715$	$SD_4 = +466.417$
$-3d_1d_2 = -1.675$	$-4d_1d_3 = -6.150$	$4d_1SD_3 = +5.855$
$2d_1^3 = .000$	$6d_1^2d_2 = +.152$	$6d_1^2SD_1^2 = +.152$
	$-3d_1^4 = .000$	$d_1^4 = .000$
$SD_3 = +33.268$	$SD_4 = 466.717$	472.724 should check with $d_4 = 472.715$

$$\frac{SD_3}{SD_1^3} = \frac{33.268}{45.195} = +.736 \qquad \frac{SD_4}{SD_1^4} - 3 = \frac{466.717}{160.985} - 3 = 2.899 - 3 = -.101$$

$$\text{Coefficient of asymmetry} = B_3 = \frac{.736}{-6} = -.123$$

$$\text{Coefficient of excess} = B_4 = \frac{-.101}{24} = -.0042$$

The several combinations of the individual "d" values lead finally to quantities which give directly the coefficients of asymmetry and excess mentioned previously. A check computation is also included which indicates whether substantial errors were made in all previous computations.

The standard errors of the computed quantities are found:

$$\text{Error of average DBH} = \frac{SD}{\sqrt{NT}} = \frac{3.562}{\sqrt{158}} = \pm .283$$

$$SD = \frac{SD}{\sqrt{2NT}} = \frac{3.562}{\sqrt{316}} = \pm .200$$

$$B_3 = \frac{1.9325}{3\sqrt{NT}} = \frac{1.9325}{3\sqrt{158}} = \pm .051$$

$$B_4 = \frac{.6124}{3\sqrt{NT}} = \frac{.6124}{3\sqrt{158}} = \pm .016$$

A word or two in explanation before proceeding further. The first of these two coefficients (B_3) defines the amount and character of the skewness, that is to say, the extent a distribution is warped from the normal and the direction in which it is warped. The second coefficient (B_4) defines to what extent a given distribution exceeds the normal at both the center and the ends of the distribution. If B_3 and B_4 equal 0 the distribution is normal. A minus B_3 indicates skewness toward the small diameters, a plus B_3 one toward the large diameters. The larger the coefficient is the more pronounced is the asymmetry. A positive B_4 indicates that the frequency exceeds the normal near the mean and at the two ends of the distribution and is less than the normal in the region between these portions of the curve. A negative B_4 indicates the opposite condition.

The formula of theoretical frequencies into which these coefficients are substituted and the solution of which is brought about by Charlier's tables is:

$$F(x) = \frac{NT}{SD}(\phi_{0(x)} + B_3\phi_{3(x)} + B_4\phi_{4(x)})$$

and the computation follows this tabulated form:

Computation of theoretical frequencies

x_1	x_2	x_3	x_4	----------
$\frac{x_1 - \text{Av. DBH}}{SD}$	$\frac{x_2 - \text{Av. DBH}}{SD}$	$\frac{x_3 - \text{Av. DBH}}{SD}$	$\frac{x_4 - \text{Av. DBH}}{SD}$	----------
$\phi_{0(1)} =$ $B_3\phi_{3(1)} =$ $B_4\phi_{4(1)} =$	For diameter x_1 in terms of Av. DBH and SD as above from Charlier's tables		$\phi_{0(4)} =$ $B_3\phi_{3(4)} =$ $B_4\phi_{4(4)} =$	----------
$\text{Sum}_1 =$	$\text{Sum}_2 =$	$\text{Sum}_3 =$	Sum_4	----------
$\frac{NT}{SD}\text{Sum}_1 =$	$\frac{NT}{SD}\text{Sum}_2 =$	$\frac{NT}{SD}\text{Sum}_3 =$	$\frac{NT}{SD}\text{Sum}_4 =$	----------

The results are the theoretically fitted values.

The first step in this stage is to present the difference between each diameter group (x_1, x_2, etc.,) and the average diameter in terms of the standard deviation $\left(\frac{x - \text{Av. DBH}}{SD}\right)$. Then from Charlier's tables with B_3, B_4 and the above known, the function values $\phi_{0(x)}$, $B_3\phi_{3(x)}$, $B_3\phi_{4(x)}$ can be read off for each diameter. These three function values are summed, multiplied by the total number of trees, and divided by the standard deviation. This gives the number of trees to be found theoretically in each diameter group.

This last stage is not applied to the plots represented in this study, since there are too few values to form a basis for reliable stand tables. However, a number of B_3 and B_4 values were computed for representative plots, and are listed to show the trends of these coefficients and hence of the diameter distribution of these plots. (Table 13.)

TABLE 13.—*Coefficients of asymmetry and excess*

Pilot	B_3				B_4			
	First	Second	Third	Fourth	First	Second	Third	Fourth
Siuslaw 4	−.023	−.020	+.003	+.005	−.030	−.026	−.019	−.017
Siuslaw 5	−.125	−.123	−.123	−.107	+.002	−.0042	+.010	+.004
Columbia 2	+0.54	+.065	+.074	----------	−.001	−.001	−.001	----------
Cascade 1	−.037	−.027	+.006	+.023	−.035	−.033	−.028	−.028

All four plots illustrate the tendency to proceed in their distribution of diameters from a negative skewness (an asymmetry toward the small diameters) on to normal form and beyond into a more positive skewness (an asymmetry toward the large diameters). The addition of more plots to form composite stand tables would make the advance more definite and would decrease the standard errors,

which have proved large for plots with so few trees and as a result have made the values of the coefficients uncertain. A coefficient value greater than three times its standard error is usually taken as a significant value.

SUMMARY

The outstanding facts brought out so far in this study of a few permanent sample plots through a period of 15 years may be briefly summarized as follows:

REGION.—Even-aged stands of pure second-growth Douglas fir are of common occurrence in Oregon and Washington west of the Cascade range.

PLOTS.—In this region in the last 15 years at various times a total of 20 permanent sample plots have been established upon which periodic growth measurements have been made. Of these, 12 have been measured more than once and the results analyzed. Three plots on the Cascade National Forest are in a Site II forest, present age 69 years, and have been under observation for 15 years. Two plots on the Siuslaw Forest are in a Site II stand, now 65 years old, and have also been under observation for 15 years. Two others on the same forest, remeasured for an equal length of time, are in a Site I stand, now 53 years old. On the Columbia five plots in an 83-year-old stand, one on Site IV and four on Site III, have been observed for 10 years.

VOLUME.—At the last measurement the Cascade plots averaged a volume per acre of approximately 11,000 cubic feet, 80,000 board feet International rule, or 53,000 board feet Scribner rule; the three existent Columbia plots average now 12,000 cubic feet, 87,000 board feet International, or 58,000 board feet Scribner; the Site II Siuslaw plots (65 years old) carry per acre 14,600 cubic feet, 103,000 or 64,000 board feet International and Scribner, respectively, and the Site I Siuslaw plots (53 years old) 10,000 cubic feet, 71,000 board feet International, or 47,000 board feet Scribner.

MORTALITY.—On an average, death takes about 2 per cent of the basal area every five years, or about 2 per cent of the cubic foot volume. Since the dead trees are chiefly of the smaller diameter this means about 1 per cent of the International board-foot volume and less than 0.5 per cent of the Scribner volume.

INCREMENTS.—The mean annual increments for cubic feet and International board feet have all neared or passed their maximum, but for Scribner volume they are still approaching it in all cases.

BOARD FOOT-CUBIC FOOT RATIOS.—Board foot-cubic foot ratios which show the average number of board feet to the cubic foot, computed by dividing the total board-foot volume of a plot by the total cubic-foot volume, are known to increase normally with increase in age. In these plots, the International ratio seems to have reached its high point. Practically all trees have passed the minimum diameter of 7 inches, and further increase can be expected to be much slower than in the early life of the stand. The Scribner ratio, since it is based upon a minimum diameter of 12 inches, still shows a decided increase, for a good proportion of the trees lie below the 12-inch minimum.

NORMALITY.—Based upon the relation of the sample-plot values to the values shown in the tables of the recently completed yield study for second-growth Douglas fir, expressed in percentages of the

first to the second, called "normality" percentages, tentative conclusions are drawn as to the approach of abnormally stocked stands to a normally stocked condition. The amount of change in normality percentage for a five-year period is different for each factor. For plots below 100 per cent the average advance in normality for a five-year period for total basal area is 2.7 per cent, for cubic foot 4.5 per cent, for International volume 6.1 per cent, for Scribner volume 2 per cent.

DIAMETER GROWTH.—On Site I average diameters increase at the rate of 1 inch in 3 years during the ages covered by these permanent plot studies, on Site II 1 inch in 5 years, on Site III 1 inch in 6 years, while on the one Site IV it takes almost 7 years for the average diameter to increase 1 inch. The diameter growth per tree increases on an average with the size of the tree and is shown graphically for several cases. Every period shows a decrease in diameter increment in each diameter class from the preceding period.

NUMBER OF TREES IN EACH DIAMETER CLASS.—The character of the curve of stem distribution in diameter classes evidently follows definite trends as the stand grows older, such as from a skewness toward small diameters to a more normal or symmetrical distribution and beyond that to a more and more positive skewness (an asymmetry to the large diameters). Double frequencies or distributions with two peaks seem to merge into one in the course of time.

A ROT OF GLADIOLUS CORMS CAUSED BY PENICILLIUM GLADIOLI, L. McC. AND THOM [1]

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INTRODUCTION

In May, 1926, a large shipment of gladiolus corms of the Odin variety arriving in the United States from the Netherlands was held up by the plant quarantine inspection service because of the generally bad condition of the corms. A casual examination showed that many were extensively rotted. Various fungi were fruiting on the surface, and mites were present. In an effort to discover what organism was mainly responsible for the trouble, examinations and cultures were made from a large number of corms in various stages of disease. The results of this study indicated that two fungi were causing most of the decay, one a Fusarium, the other a Penicillium. The latter is the subject of this paper.

The corms having the Penicillium type of rot had rather firm but not hard, reddish brown, sunken spots, sometimes slightly roughened by irregularly concentric wrinkles, more or less irregular in size and shape, margins indefinitely blending with the normal epidermis. Some lesions had no visible surface growth; others had a scanty growth of white mycelium and small sclerotia. Sections of such lesions showed the brown rot extending deeply and without definite margins into the flesh of the corms. The texture of the rotted tissues was only slightly porous, moderately firm and dry, but never dense and hard as in "hard rot." With a lens some white mycelium was seen in small cavities. In some of the corms sclerotia were found embedded in the tissues.

These sclerotia were smooth and cream colored, but in contrast to the brown tissues by which they were rather closely surrounded they appeared white. The sclerotia were found only in the older parts of the lesions. (Pl. 1, E.) Other corms with the same type of lesions had no sclerotia.

For cultures, bits of tissue were taken from the inner margin of lesions on corms with and without sclerotia. All of the cultures produced the same type of growth in which numerous sclerotia developed. The cultures seemed contaminated with a Penicillium, and numerous unsuccessful attempts were made to separate the sclerotia-forming fungus from the Penicillium. Sclerotia were isolated, sterilized by dipping in alcohol to remove air, then in mercuric chloride, 1:1,000, for periods up to 20 minutes. After being rinsed in sterile water, some of the sclerotia were placed at once on culture media; others were kept under dry conditions for several weeks, then put on culture media. Isolated single hyphae tips were used and also single conidia of the Penicillium. All of these sources of inoculum regularly and persistently produced both Penicillium conidia and sclerotia.

[1] Received for publication Nov. 14, 1927; issued April, 1928.

In June, 1926, the same type of lesion was found on gladiolus corms grown in New Mexico, and cultures from these produced Penicillium and sclerotia with the same characteristics as those from the Netherlands corms. Very shortly afterwards the same type of growth was also isolated from diseased Tigridia bulbs grown in New York. The gladiolus corms from New Mexico and the Tigridia bulbs were in more advanced stages of decay than the Netherlands corms, and both lots of material had numerous surface as well as interior sclerotia.

Another series of experiments for separation of the Penicillium and the sclerotia resulted in failure; so finally it seemed quite certain that the two forms belonged to the same fungus.

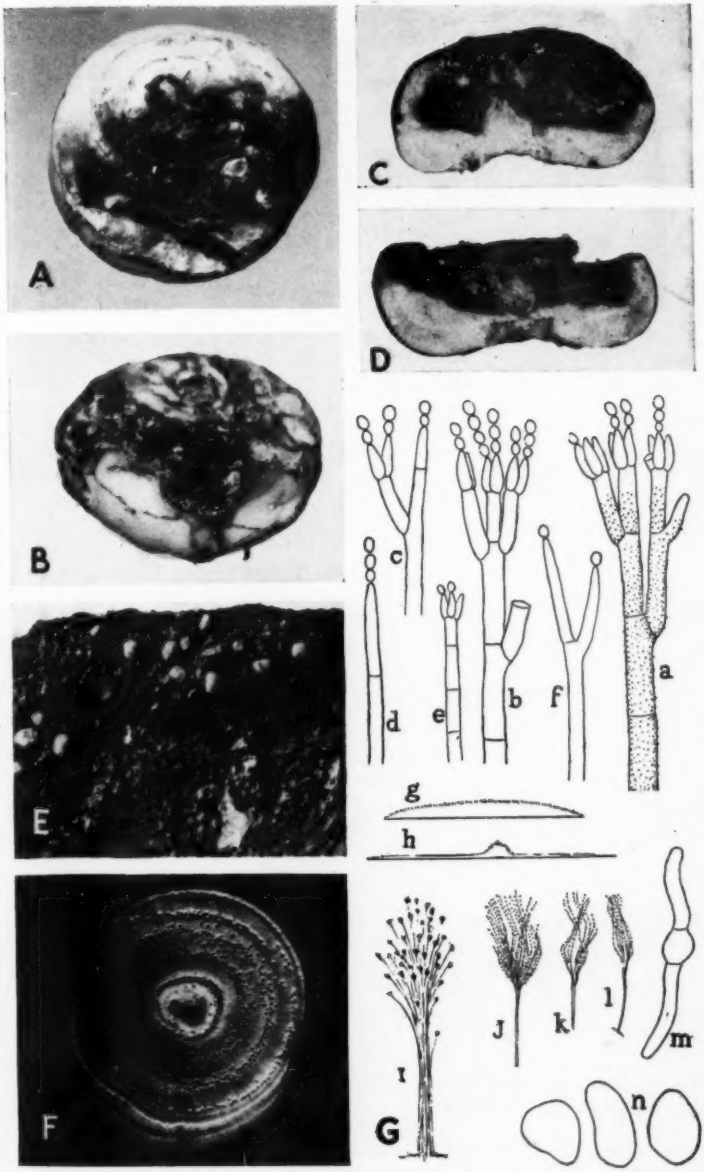
INOCULATION EXPERIMENTS

A number of perfectly clean gladiolus corms of the varieties Peace and America were husked, dipped in alcohol, covered with mercuric chloride for 10 minutes, rinsed well in sterile water, and then placed separately in sterile, covered dishes. The corms were inoculated with pure cultures of the fungus, using young mycelium not yet producing sclerotia for some, and for others older cultures producing sclerotia. One-half of the corms had the inoculum placed on the intact epidermis. The other half had small wounds made with a sterile scalpel and the inoculum placed over the injured tissue. After five weeks at 22°–24° C. the fungus had failed to penetrate the epidermis of the unwounded corms. The wounded corms, on the contrary, soon showed a slight growth about the wounds and a discoloration which spread out fairly rapidly over the surface. This discoloration was a dull brown in color (Dresden brown and umber to sepia in Ridgway's colors) (*4*).[2] There was never a luxuriant growth on the surface of the corms, and only a few tufts of the dull gray-green Penicillium developed about the wounded areas. (Pl. 1, A, B.) Sections of these corms showed considerable penetration of the decay (pl. 1, C, D) and numerous sclerotia scattered through the dark-brown tissue. The Netherlands, New Mexico, and Tigridia strains of the fungus gave identical results.

Gladioli growing in pots were copiously inoculated by placing young cultures and sclerotia about the stems and directly on the growing corms. No wounds were made. These plants when mature, two months later, were entirely normal. A few inconspicuous-brown spots were found on various parts of some of the plants, but the Penicillium was never recovered in any of the cultures made from these spots.

In another experiment on gladioli in pots, opposite sides of large growing corms were inoculated. A section of the husk was carefully lifted and the inoculum placed directly on the epidermis. One side of each corm was wounded by pricking with a sterile needle, the husks were then replaced, and the corms re-covered with soil. Four weeks later, half of the corms were dug and examined. All the wounded areas showed infection as dark-brown spots with water-soaked margins. This dark-colored rot had penetrated 4–8 mm. into the flesh of the corm. Six weeks after inoculation the remaining corms were dug, and only the wounded sides showed infection. From the dark brown interior tissues of these inoculated corms the fungus was isolated in pure culture.

[2] Reference is made by number (italic) to "Literature cited," p. 224.



A, B, C, D.—Gladiolus corms inoculated with *P. gladioli*, four weeks after inoculation. × 1
E.—Sclerotia in corm tissues. × 8
F.—Thaxter agar colony. × 1
G.—*a*, Conidiophore from Czapek agar culture grown at 15° C., × about 650; *b*, grown at 22° C., × about 650; *c*, *d*, *e*, *f*, conidiophores in cultures grown at room temperature, × about 650; *g*, cross section of colony grown at 15° C., × ½; *h*, colony grown at 22° C., × ½; *i*, coremia, × 10; *j*, *k*, *l*, penicillia, × 45; *m*, germinating conidium, × about 650; *n*, sclerotia, × 25. From camera lucida drawings

Tigridia bulbs were not used in any of the inoculation experiments' but the fungus isolated from the diseased Tigridia bulbs is culturally like that from the Netherlands and the New Mexico corms and it produces the same sort of decay and sclerotia when gladiolus corms are inoculated with it.

From these experiments it appears probable that this fungus is not dangerous except to wounded corms or to corms kept under very unfavorable conditions.

CULTURAL CHARACTERS

At average room temperatures (22°–24° C.) the fungus presents an entirely different aspect from that shown when grown at 14°–15°. At the higher temperature the mycelial and conidial development is more or less suppressed and the abundant sclerotia are the conspicuous feature of the growth. At the lower temperature the luxuriant growth of white mycelium and green conidia delays and often hinders the production of sclerotia.

Czapek's solution agar and potato-dextrose agar are favorable media for this fungus. Single sclerotia placed on plates produce in three days at room temperature areas of growth 3–5 mm. in diameter. The growth is dense and pure white except at the water-soaked margins. Sclerotia begin to develop about the sixth day in a zone surrounding the center of growth and continue to develop in successive zones as long as the colony grows. (Pl. 1, F.) As the sclerotia develop, the mycelium becomes thin and inconspicuous. In the areas where sclerotia are growing, the hyphae bear numerous drops of clear, pale orange-yellow liquid. Immediately following the production of the first sclerotia or at the same time there develops in the center of the colony a small tuft of the dull gray-green (mineral gray to tea green) (*5*) Penicillium. Colonies 40–60 mm. in diameter have Penicillium tufts 2–4 mm. in diameter. The reverse side of the growth when 2 to 3 weeks old is reddish brown (Sudan brown) (*5*) in the Thaxter agar plates and light pinkish cinnamon in the Czapek agar plates. In crowded plates where the colony growth is limited to 10 mm. or less the central tuft of Penicillium does not develop sufficiently to be visible, but conidia and conidiophores can be found by microscopic examination.

In tube cultures the growth is more luxuriant than on plates, and by the tenth day the numerous sclerotia form a continuous layer over the surface of the slant, the white mycelium showing scantily at margins and between sclerotia. Penicillium tufts may or may not be visible (conidiophores and conidia, sometimes small and imperfectly formed, can be found in a microscopic examination), and the drops of clear liquid are abundant.

Finally the white mycelium becomes scanty and inconspicuous, leaving the coarse, granular layer of closely packed sclerotia, with perhaps a small central area of dull green.

The individual sclerotia are palest yellow or a cream color, but in mass as seen in cultures they are pale pinkish tan. Light drab and drab are the closest matches found in Ridgway (*5*), but these are scarcely pink enough.

When grown at 14°–15° C. there is a luxuriant mycelial growth, and the whole surface is soon entirely covered with the bluish gray-

green conidia. The conidiophores bear large, definite heads, and coremia 1–4 mm. tall develop. Sclerotia are slow (three to five weeks) in forming, and in shallow layers of agar they often fail to develop. The conidial areas gradually lose their blue-green color, becoming olive gray and later dark brown, sepia to mummy brown (*5*).

On oatmeal, potato, malt, wort, oxalic acid, and whey agars, the growth is similar to that on potato-dextrose and Czapek agar. Shrinkage of the media often causes the growth to fall into irregular, wide folds. The medium is not discolored, except for a slight browning of beef agar and gelatin. The reverse colors of the growth vary from cinnamon buff to Sudan brown. The cultures have no odor.

On beef agar the growth is scanty, at first white but finally a cream color. The reverse color is yellow to reddish brown. There is a scanty development of conidia and few to no sclerotia.

Nutrient beef gelatin at 22° C. produces a moderate amount of white surface growth, with a bright reddish brown substratum. Liquefaction is very slow. The addition of 1 per cent saccharose did not improve the growth or hasten liquefaction.

Plain gelatin (15 per cent) in tap water produced very scanty growth and no liquefaction in two months.

Lactose agar (synthetic base and colored with brom cresol purple) produced only moderate mycelial growth but numerous sclerotia. The conidial fructification was a brighter green (celandine green to artemisia green) (*5*) than on most culture media. The reaction was acid.

Litmus milk cultures first redden the cream rim. Whey forms on the surface, but there is no evidence of curd. The clearing gradually extends downward and is complete in two weeks at 22°–23° C., the whole of the medium becoming translucent and a dull wine color.

Sterilized gladiolus corms were cut in half and inoculated with the fungus. Half were kept at 14° C. and half at 25°–26°. At the lower temperature the corms in three to four weeks were covered with a blue-green layer of conidia, which persisted for weeks, while at the higher temperature only scanty white mycelium, short and dense, developed. The gladiolus tissues were completely destroyed in two months, and sclerotia were produced in large numbers at both temperatures.

Ripe oranges were pricked with a sterile needle and inoculated with the fungus. A dark brown, only moderately soft, rot developed. After one month at room temperature the rot had spread from the small inoculated area throughout the whole orange. The rind was dark brown, but there was no surface growth of fungus except at the inoculated point, where there were a few gray-green tufts and some small, white, immature sclerotia. At 14° C. the rind was less dark in color and there was a luxuriant surface growth of bluish gray-green conidia on isolated conidiophores and coremia and also a number of immature white sclerotia. Examination showed that at both temperatures the mycelium had penetrated to all parts of the interior, but without causing any particular change in the appearance or odor of the oranges. Sclerotia were very abundant in the tissues of the rind. The fungus did not penetrate uninjured orange epidermis.

On most of these different media, cultures were grown at room temperature and also at 14°–15° C. At the lower temperatures the conidial growth is definite and usually abundant, while at the higher

temperatures conidial production is scanty and in some cases almost absent. Beef medium at room temperature has very few and very poorly developed conidiophores. (Pl. 1, G, *d*, *e*, *f*.) However, at room temperature the chains of spores are longer and resist breaking up better than those formed at 14°–15°. In favorable media at the lower temperature the conidiophores are taller and coarser and more branched than at room temperature, and the walls are finely roughened. (Pl. 1, G, *a*.) At the higher temperatures the same media produce smooth-walled conidiophores. (Pl. 1, G, *b*, *c*.)

TEMPERATURE RELATIONS

Parallel plate cultures of potato-dextrose agar were made and kept at 14°, 22°–23°, and 30° C. At the lower temperature growth is slower than at 22°–23°, but, continuing for a longer period, eventually covers more surface, producing dense mycelium and abundant conidia not only in the center of the medium but over all but the extreme margin. Sclerotia did not develop until after three weeks.

In three weeks at 22°–23° C. the medium was drying out and the thin creamy-white growth was thickly sprinkled with numerous sclerotia, the central area of gray-green being 3–5 mm. in diameter. At 30° a few hyphae developed on the bits of inoculum, but growth never spread out until after three weeks, when the temperature dropped to 27° and a scanty growth pushed out into the surrounding medium. Tube cultures grown at the same temperatures gave comparable results.

Sterilized gladiolus corm tissues were inoculated with bits of mycelium and grown at 5°, 10°, 15°, 20°, 25°, and 27° C. Five days after the inoculations there was at 5° no visible growth; at 10° very scanty white growth; at 15° a moderate amount of white mycelium and a small green area; at 20° better growth and more green color than at 15°; at 25° less growth and less green than at 15° and 20°, sclerotia present; at 27° no visible mycelium, no green color, sclerotia present.

In 15 days at 5° C. there were scanty white mycelium, no conidia, no sclerotia, and no discoloration of the medium. At 10° all exposed surfaces of the medium were covered with growth, bluish gray-green with pure white margins; no sclerotia present; medium not discolored; conidiophores in coremia 1–2 mm. tall, columns not very definite, white or colorless; only a trace of roughness on walls of the conidiophores. At 15° conidial development was more luxuriant than at 10°; numerous drops of pale yellow to reddish orange liquid; coremia 2–4 mm. tall, lower half of the columns pinkish to orange red; sclerotia numerous; conidiophores definitely rough walled; medium discolored and porous. At 20° there was less conidial development than at 10°, a few poorly developed coremia present; mycelium not visible; all exposed surfaces covered thickly with sclerotia; walls of conidiophores only slightly or not at all rough; medium discolored and some sclerotia forming in the parts just below the surface growth. At 25° surfaces were covered with sclerotia, scanty green color, but well-developed conidiophores were found. Evidently the gladiolus tissue is a more suitable food than any of the artificial media, which produced very poor conidiophores at 25°. At 27° a short dense layer of dull white mycelium and small sclerotia were observed, covering about one-

fourth of the exposed medium; there was no green color; no conidiophores were found in microscopic examination; the medium was discolored and porous.

VITALITY

Potato-dextrose agar tube cultures, kept at room temperature and becoming rather dry, were alive at the end of 10 months, but were evidently reduced in vitality, for quite a number of the 10-month-old cultures failed to develop growth when transferred to fresh media. Cultures 6 to 9 months old readily renewed growth when transferred to fresh media.

Sclerotia, sterilized in 50 per cent alcohol for 2 minutes, then in $HgCl_2$ 1–1,000 for 15 minutes, were slow in renewing growth. Several started growth after the thirteenth day, and one remained dormant 40 days before showing signs of life. Sclerotia sterilized 20 minutes in $HgCl_2$ 1–1,000 showed the first growth 9 days later; by the twelfth day all were growing.

Sclerotia taken from dry corms, sterilized in alcohol two minutes, in $HgCl_2$ 1–1,000 for three minutes, rinsed well, and placed on favorable media, promptly produced growth. Some of these sterilized sclerotia were kept dry in a tube for 12 weeks, then put on media, but none of these produced growth.

Sclerotia from diseased corms that had been stored at room temperature for 11 to 12 months did not grow when placed in favorable media.

Dry sclerotia treated with 0.25 per cent aqueous solutions of Uspulun and Semesan up to 45 minutes, then rinsed several times in water, produced growth. Similar treatment was given to mature but still moist sclerotia, with the same results. Dry sclerotia treated with mercuric chloride 0.1 per cent (1–1,000) in water for 15 minutes were mostly killed. After 30 minutes' treatment no growth resulted. But moist sclerotia were much less affected by the same treatment. Growth was delayed but not destroyed by even 45 minutes' treatment with the mercuric chloride. This difficulty in killing the fungus, even when it is possible to apply the fungicide directly to the sclerotia, indicates the probable lack of success of control measures based on the sterilization of infected corms.

SOME OTHER SCLEROTIAL DISEASES

Several other known diseases of gladioli are caused by sclerotia-forming fungi. The dry-rot disease described by Drayton (*2*) is apparently definitely pathogenic to growing plants. No spore stage has been found for this fungus. The sclerotia are black and much smaller than those of *Penicillium gladioli* L. McC. and Thom.

Aspergillus ochraceus Wilhelm (*7, 9*) has been isolated by the senior writer from diseased gladiolus corms. The sclerotia formed by this fungus are larger and darker than those of *Penicillium gladioli*, and they are always accompanied by the typical spore stage of the Aspergillus.

From diseased Ixia corms the senior writer isolated a fungus which causes rapid and extensive rot of gladiolus corms under storage conditions. Numerous large dark-brown sclerotia develop, but no spore stage has been found. The identity of this fungus has not been determined. It may be identical with *Sclerotium tuliparum* Klebahn (*3*).

Another fungous disease of gladioli producing black sclerotia has been described by Voglino (*8*) and named *Sclerotinia bulborum*.

Schwartz (*6*) has reported a sclerotium-producing Penicillium which he and Wehmer identify as *P. italicum*. There seemed a possibility that the Penicillium of the gladiolus corms might be the species described by Schwartz. However, a study of a culture of this particular *P. italicum*, obtained from Schwartz, shows that it is culturally and morphologically unlike *P. gladioli*.

TECHNICAL DESCRIPTION[3]

PENICILLIUM GLADIOLI

Colonies presenting two aspects: When grown at temperatures above 20° C., predominantly consisting of sclerotia with few and inconspicuous conidiophores; when grown at 15° C. or lower, showing abundant green conidial areas with delayed or partially suppressed sclerotium formation. Upon Czapek's solution agar at 20° to 24° C. producing a thin aerial felt of mycelium with sclerotia beginning about the sixth day and developing in successive concentric zones, and giving the characteristic appearance of the species; sclerotia 140μ to 540μ in diameter, at first cream to light pinkish tan, in age very pale brown or tan, smooth, and composed of thick-walled cells 8μ to 12μ in diameter, retaining their vitality several months; reverse light pinkish cinnamon; drops of orange-yellow fluid more or less conspicuous; odor none, conidiophores few, scattered and inconspicuous among the sclerotia, often very long (up to 2 mm.) and about 2μ to 3.6μ in diameter, later developing in more or less conspicuous tufts, fascicles, or complex branching coremia in the center of the colony and definitely green (bluish gray-green); penicillus consisting of the main axis of the conidiophore with or without one or two branches, bearing few metulae 10μ to 12μ long and verticils of few sterigmata 12μ to 14μ by 1.5μ to 2μ with tapering rather than acute points, and conidia elliptical-fusiform, smooth, hyaline, 2.8μ to 3.6μ by 2.5μ to 3μ, adhering in long chains in fluid mounts, more or less parallel then tangled as seen in the penicillus; swelling to 6μ or 7μ in diameter and germinating by one or two tubes.

Colonies grown at 14° to 15° C., producing abundant mycelium and conidial areas gray-green (light dull glaucous blue, glaucous blue, and greenish glaucous blue of Ridgway (*5*)), to the very margin of the colony; sclerotium formation delayed and reduced, not dominating the growth; conidiophores partly simple, partly aggregated into coremia, tending to be longer and coarser, and when in coremia commonly 3μ to 4.5μ or even 6μ in diameter with walls pitted or roughened; pencillus coarser, more branched, and with larger verticils of sterigmata and the elements varying in diameter but following the conidiophore in being coarser than in the colonies grown at higher temperature; conidia not different from the conidia produced at higher temperature.

The conidial form described and figured here complies closely with the description of *Penicillium divergens* of Bainier and Sartory (*1*) and would have been identified as that species except for the entire lack of sclerotia in that species.

Found as a cause of decay in gladiolus corms. Specimens of natural and artificial infections on gladiolus corms and dry cultures have been deposited in the herbarium of the Office of Mycology and Disease Survey, Bureau of Plant Industry, United States Department of Agriculture.

SUMMARY

A disease of gladiolus corms and Tigridia bulbs caused by *Penicillium gladioli* is described. As known at present it is chiefly a storage disease. Experiments have proved that the fungus under ordinary conditions does not penetrate the normal uninjured epidermis of gladiolus corms, but through even slight wounds it rapidly invades the inner tissues of both mature and growing corms, causing a dark brown, moderately dense rot. Numerous small white to cream-colored sclerotia are usually formed in the rotted tissues.

[3] This description embodies and supplements that given by McCulloch and Thom in an earlier publication (*4*).

The fungus invades the tissues with about equal rapidity at temperatures ranging from 15° to 23° C. At the lower temperatures there is abundant blue-green conidial growth and delayed formation of sclerotia, while at the higher temperatures there is scanty or no visible conidial development, but a rapid and abundant production of sclerotia.

Experiments with mercuric chloride and commercial fungicides show that the sclerotia are rather resistant.

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LILAC BLIGHT IN THE UNITED STATES[1]

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INTRODUCTION

In the summer of 1925 some blighted lilac twigs were sent to the pathological laboratory of the Bureau of Plant Industry by H. W. Anderson, of Urbana, Ill., with the inquiry whether this was the bacterial disease described by Van Hall from the Netherlands and hitherto unreported in the United States. The twigs were blackened wholly or on one side only; cross sections showed the discoloration extending sometimes only through the cortex but often involving the entire thickness of the stem. The discolored tissues of the cortex swarmed with bacteria. Poured plates yielded practically only one type of colony. This was round, white, and caused green fluorescence in the agar, but its most striking characteristic was a wrinkling of the surface, which began on the second day and was persistent. As no mention was made of this very noticeable character by Van Hall (*2*) [2] or by E. F. Smith in his unpublished notes it seemed unlikely that this was the organism with which they had worked. Good infections, however, were obtained with subcultures from these wrinkled colonies. Since no culture of *Bacterium syringae* (Van Hall) EFS. (*Pseudomonas syringae* Van Hall) was available for comparison, cultural work was carried on and the cultures compared with the brief published and unpublished descriptions of *Bact. syringae*. The results were inconclusive, however, although there was no substantial disagreement aside from the colony character mentioned above.

In the spring of 1926, Westerdijk, of the Netherlands, sent the writer some blighted lilac twigs from which the causal bacterium was readily and repeatedly isolated. This organism was proved infectious by inoculations on lilac twigs. None of the colonies in any of the isolations and reisolations from this material showed any trace of wrinkling.

Cultural comparisons of the Netherlands and Illinois isolations were then made, but no real differences aside from that mentioned were found. The effects produced by inoculation with the two strains likewise showed no distinguishable differences. (Fig. 1, A and B.) It seems evident, therefore, that the two are strains of the same organism (*Bacterium syringae*) and that the Illinois disease is identical with the Netherlands disease.

While the work here reported was in progress a note appeared by C. O. Smith (*7*) in which he states that *Bacterium citriputeale*, the cause of citrus blast and black pit, is also probably responsible for a new lilac blight in California. To quote: "This investigation is

[1] Received for publication Oct. 3, 1927; issued April, 1928.

[2] Reference is made by number (italic) to "Literature cited," p. 235.

well in progress, and the results thus far indicate that the citrus blast, the avocado blemish, and the California lilac disease are closely related and probably are caused by the same organism." Com-

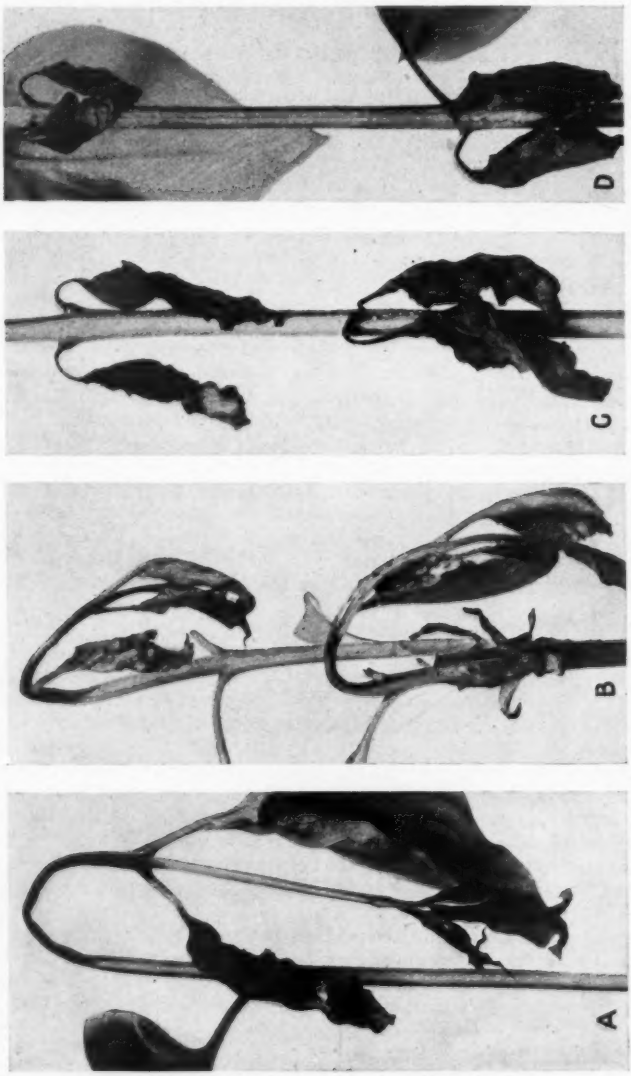


FIG. 1.—A, Lilac twig 12 days after prick inoculation with *Bacterium syringae* from the Netherlands; B, lilac twig 10 days after prick inoculation with *Bact. syringae* from Illinois; C, lilac leaves inoculated in the petioles with *Bact. syringae* from Illinois; D, lilac leaves inoculated in the petioles with *Bact. citriputeale*

parisons were therefore made between *Bact. syringae* isolated from Illinois and Netherlands lilacs with a freshly isolated culture of *Bact. citriputeale* received from Fawcett in California. Parallel cultures proved the latter to be identical with *Bact. syringae* except

in colony characters on agar, and the differences in these were no more striking than the differences between the Netherlands and the Illinois strains. Inoculations on lilac with *Bact. citriputeale* gave good typical infections. (Fig. 1, D.) Good black-pit lesions were also produced on lemons by needle pricks with both the Netherlands and the Illinois strain; but these strains were somewhat less virulent on lemons than *Bact. citriputeale.*

From these comparisons it seems probable that the lilac blight of California is identical with that in Illinois and the Netherlands.

HISTORY OF THE DISEASE

Lilac blight has heretofore been reported only from the Netherlands, Germany, and England.

It was first observed by Sorauer (*9*) in Germany in 1891. In 1899 Ritzema Bos (*5*) found it in the Netherlands. Both considered it to be bacterial. Beyerinck subsequently obtained infections on lilacs with an organism which he isolated and studied. He did not publish, but turned over the organism and the results of his inoculations to Van Hall (*2*) who in 1902 published a detailed account of the appearance and progress of the disease. Van Hall made cultural studies of the bacterium, which he named *Pseudomonas syringae*, although he did not succeed in producing any infections with his organism.

Güssow (*1*) in 1908 published his observations on a lilac blight in England which he ascribed to *Pseudomonas syringae.*

In 1906 E. F. Smith while in the Netherlands studied this blight, made observations on its effect on the plant, and obtained good infections with the bacterium which he isolated, confirming Van Hall's work. He did not, however, publish the results of his studies.

APPEARANCE OF DISEASED PLANTS

As described by Van Hall (*2*), Sorauer (*9*), and E. F. Smith, the disease in Europe is identical in appearance and development with that in Illinois. Güssow's (*1*) description of the blight in England does not, however, conform to these descriptions. He does not mention stem or petiole infections, but confines his account to the appearance of the leaf blades, and even here the symptoms are unlike those of the Netherlands disease. His reported infections on leaf blades from needle pricks did not appear until two months after inoculation, while *Bacterium syringae* gives evidence of infection by the third day. It seems quite improbable, therefore, that he was dealing with the lilac blight of the Netherlands.

On the specimens received from the Netherlands in 1926 there were very distinct dark water-soaked angular spots on some leaves, usually on those with infected petioles. These spots swarmed with bacteria and yielded pure cultures of *Bacterium syringae* on plating. There were also lesions on the peduncles and pedicels of the flower clusters, and many flower buds were completely blackened.

In 1925, when Anderson first observed the disease in a large planting of lilacs in an arboretum in Illinois, their appearance suggested pear blight. Entire young shoots were blackened, some were bent, others erect. In some cases only one side of the shoot was involved, a discolored streak from several millimeters to one-half the circum-

ference of the stem in width extending the entire length of the season's growth. No definite leaf spots were observed, but leaves on the infected parts invariably were partially or entirely blackened.

In 1926 the disease did not appear at all in the planting of lilacs where it was found the previous year (see "Control," p. 232), but a less virulent outbreak occurred in the same arboretum on young nursery stock at a considerable distance from this planting. The season had been very dry, and when the plants were observed by the writer in July only small stem lesions from 5 to 15 mm. long could be found. Above and below these lesions, however, the leaves were rather thickly peppered with small irregular to round brown spots surrounded by a yellowish halo. (Fig. 2, A.) On the leaves nearest the stem lesion the leaf blades were ragged with large brown areas, due to coalescing spots and the torsion caused by irregular development of healthy and diseased areas. (Fig. 2, A.) The smaller spots were identical in appearance with those produced by inoculation in Washington. (Fig. 2, B and C.) (See "Inoculations.")

The disease attacks the young shoots as they develop in early spring, the first symptoms being brown spots on leaves and internodes. These become black and increase rapidly in size, especially in rainy weather. Further development depends largely on the age of the part attacked. On somewhat mature stems the spots usually enlarge longitudinally, passing over to the petioles in their path, causing the death of these leaves, but not of the remainder of the twig, which remains upright. When the spots occur on very immature internodes, on the other hand, the infection spreads around the twig, girdling it completely for a distance of several centimeters. (Fig. 1, A and B.) The stem bends over at this lesion, and the part above it withers and dies.

Similar differences are observable in the results of infection on immature and mature leaves. The immature leaves blacken rapidly and completely, while on older ones the spots enlarge slowly and at the worst, by coalescing, blacken and kill large areas of the blade, causing distortion.

TISSUES INVOLVED

The disease is primarily one of the parenchyma, although the vascular region may become involved. In the stem the bacteria spread through the intercellular spaces of the cortex, causing blackening and collapse of the cells and frequently cavities of greater or less extent. If the infection reaches the vascular system it may run up through this channel without external lesions, evidencing itself first by a wilting of the upper leaves, later by sunken blackened areas on the stem along the line of internal infection. In such cases, cavities filled with bacteria are formed in the vascular region. In infected petioles the parenchyma is very susceptible. The whole thickness of the petiole blackens; the bacteria make their way into the vascular system and are to be found far up in the midrib and veins, while the leaf blade blackens.

INOCULATIONS

Prick inoculations were made in the tender upper part of young lilac suckers in August, 1925, using subcultures from colonies isolated from the Illinois lilac twigs. In three days the stem near the pricks

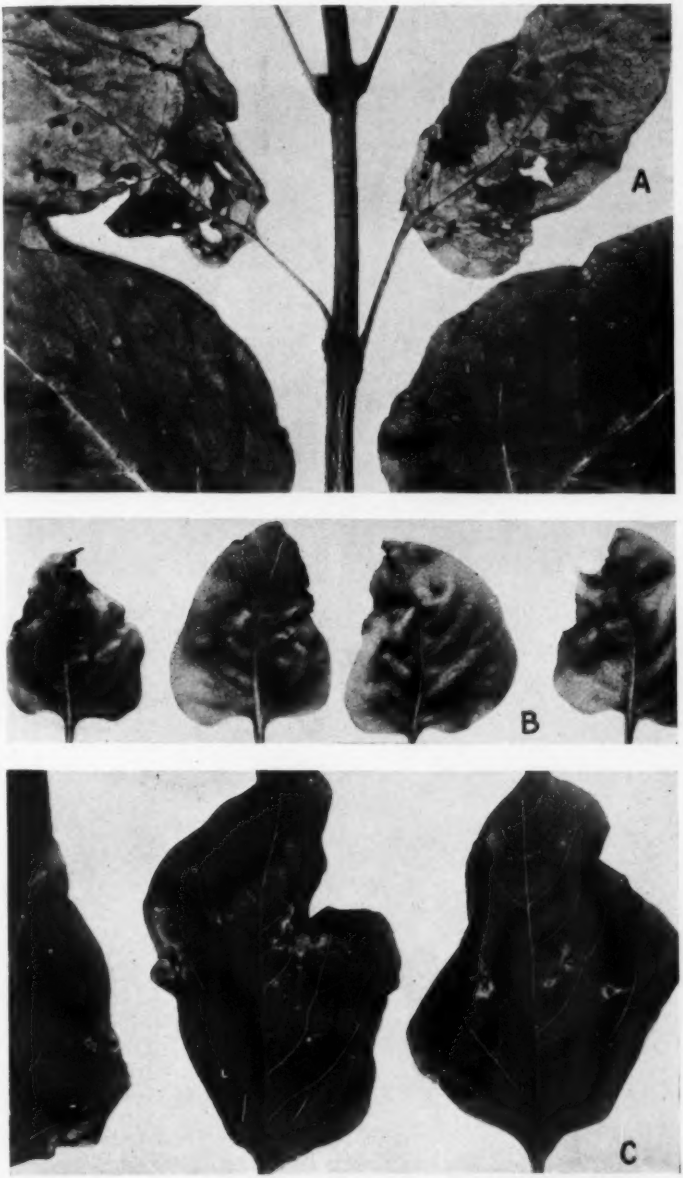


FIG. 2.—A, Natural infection on lilac leaves from Illinois, a small canker on the stem below the most seriously spotted leaves; B, lilac leaves 12 days after spraying with *Bacterium syringae* from Illinois; C, lilac leaves 14 days after spraying with *Bact. syringae* from Illinois

was blackened and the darkened area extended up into adjacent petioles. On the fourth day most of the inoculated shoots were completely dead and black above the point of inoculation. (Fig. 1, B.)

FIG. 3.—A, Infected flower clusters (the blackened drooping ones) inoculated by rubbing with *Bacterium syringae* from Illinois; B, lilac twig inoculated at X by needle pricks with Illinois strain of *Bact. syringae*; time, 16 days. The infection is advancing in the vascular region, showing in the surface lesions in two places and in the dying margins of the leaves

Isolations were made from one of these infections, and on August 25 subcultures were used to inoculate lilac shoots. Again infection was evident in two days, and after five days a dark streak extended downward through one or two internodes, invading the petioles in its

path but not causing a general blackening. Whether the whole twig blackened, or only a streak, seemed to depend largely on the age of the tissue inoculated. Pricks in the very tender tip always killed the twig; pricks made in slightly more mature wood gave only streaks. Sometimes infection spread in the cortex; in other cases it reached the vascular region quickly and spread rapidly up the stem through the vessels, as shown by the wilting of leaves in its path. Only later did a sunken black streak form externally along the line of internal infection. (Fig. 3, B.) Petioles were very susceptible, but infection did not often run back into the stem from them. When petioles became infected the whole leaf blade blackened, sometimes suddenly, at other times gradually.

The organism from the Netherlands, when used to inoculate lilacs in the pathological greenhouse, gave results identical with those obtained when the Illinois organism was used, except that the Netherlands organism seemed slightly more virulent. (Fig. 1, A.)

Stomatal infections on leaves were obtained by spraying and by rubbing with the Illinois organism and also by rubbing with the Netherlands isolations. Spots were round to irregular, brown, with a yellowish halo 1 or 2 mm. in width. The first evidence of infection was visible in five days, but the spots enlarged very slowly, causing slight distortion, and did not attain any considerable size under the very dry conditions prevailing during the lilac growing season. There was never any general spread of the disease from such infections. However, in the late fall of 1926, when rub inoculations were made on very tender leaves of young suckers during a period of rainy weather, infection spread so rapidly that individual stomatal infections were not observed. Leaves and petioles were rapidly and completely blackened. On most inoculated suckers there were also stem leasions 10 to 15 mm. long, black and sunken, and not continuous with diseased petioles, but apparently secondary stomatal infections. Isolations from these stem lesions gave wrinkled colonies from the stems inoculated with the Illinois organism and smooth colonies from those inoculated with the Netherlands organism.

SUSCEPTIBLE VARIETIES

The first observation of the disease in Illinois was on *Syringa vulgaris* only. In the nursery, however, it occurred on both purple and white sorts, *S. vulgaris* and the following varieties showing infection: Mont Blanc, Reine Elizabeth, Princess Alexandra, and Roi Albert.

Sorauer (*9*) reported that *Syringa chinensis* was less susceptible than other sorts, and Ritzema Bos (*5*) observed that *S. vulgaris* was more susceptible than *S. persica*.

ORIGIN OF THE DISEASE

An attempt to trace the infections in Illinois to importations failed, although there can be little doubt that the infection has at some time come to this country from Europe on nursery stock. Scions for the nursery where it was found came from several widely separated sources in the United States. No importations for this arboretum had been made from Europe except a few plants from France which had never been near either of the places in the arbore-

tum where infections were found, and none of the French importations showed the disease. Moreover, the disease is not known to exist in France.

CONTROL

From the nature of the disease it is evident that sprays would be of little value and that destruction of diseased shoots is the only practical remedy. Careful cutting out was used in Illinois where the disease was severe in 1925, and there was no recurrence in 1926. The unusually dry season, however, may have been a factor in the completeness of its disappearance, since the disease elsewhere was very mild in its effects.

THE ORGANISM

CULTURAL CHARACTERS

Unless otherwise stated, all beef media used were made with beef infusion and had a P_H of 6.8 to 7.2.

AGAR PLATES.—On peptone beef-infusion agar, colonies of *Bacterium syringae* Netherlands are visible in 24 to 48 hours. These are round, white, smooth, convex, finely crosshatched, and opalescent by oblique transmitted light, transparent, attaining in six or seven days a width of 5 mm. and becoming flat, but otherwise retaining all of the characters mentioned above. There is no surface wrinkling at any stage. (Pl. 1, B, D, F, and G.) The agar becomes green.

Colonies of *Bacterium syringae* Illinois, on the same agar are visible in 24 to 48 hours and at this time are indistinguishable from colonies of the Netherlands strain. On the day after they appear the center has become sunken and holds one to several clear droplets. (Pl. 1, C.) By the third day the center is deeply wrinkled, these wrinkles increasing with the growth of the colony, while the margin remains raised, smooth, and finely crosshatched. Colonies may attain a diameter of 5 mm. by the sixth and seventh day. (Pl. 1, E and H.) The agar becomes green on all beef-infusion media.

Very occasionally in direct isolations of the Illinois organism, but frequently in plates from cultures, a very few smooth colonies appear; that is, some colonies retain the smooth surface of the first day and never show any wrinkling. These when used for further plating usually give only smooth colonies, but occasionally a few wrinkled ones appear.

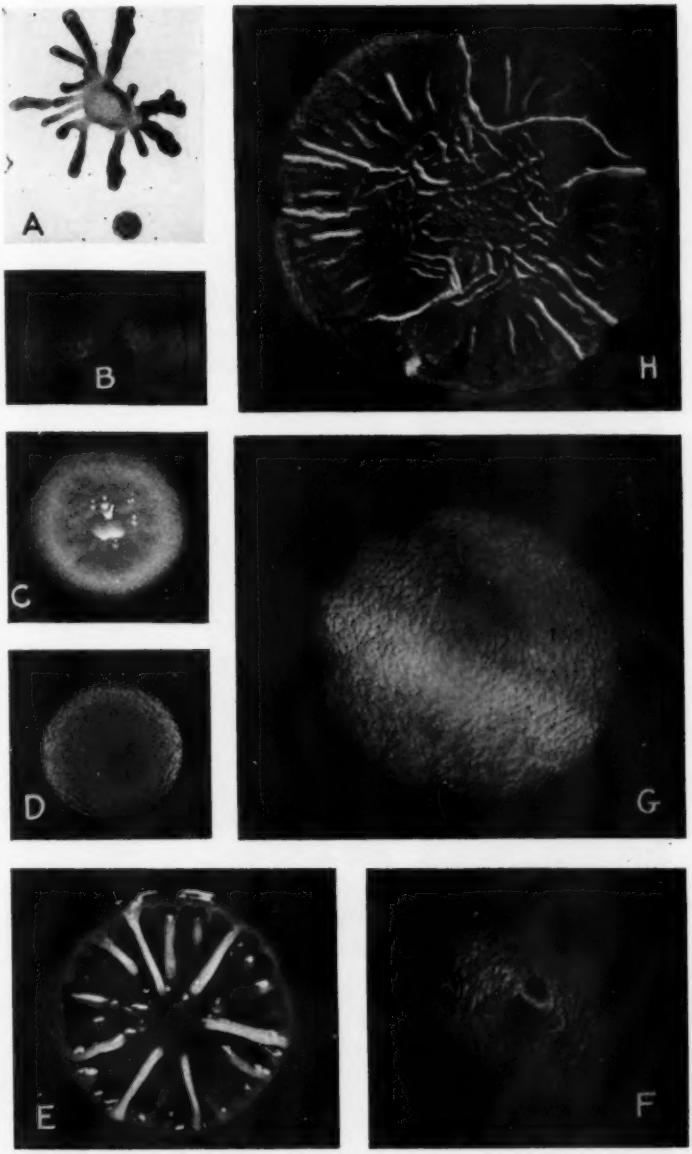
Rough and smooth strains of the same organism seem to be common to many organisms. The interconvertibility of the two types has recently been reported by Jordan (*3*) for certain animal pathogenes and by Sharp (*6*) for *Bacterium phaseoli sojense*. Rough colonies are not unusual in plates of smooth-colonied plant pathogenes. In this case the preponderance in the number of rough colonies is the unusual factor.

Occasionally spreading fimbriate colonies appeared on plates of the Illinois organism (pl. 2, A), never on those of the Netherlands organism. These colonies occasionally remained smooth but mostly became wrinkled. (Pl. 1, A.) Their occurrence seemed to depend on the surface tension of the agar, for they generally appeared on thicker portions of agar, due to an uneven bottom in the plate, and could be induced by using 20 c. c. instead of 10 c. c. of agar for a plate. This could not be done with the Netherlands strain.

Occasionally colonies very irregular in outline, slightly roughened on the surface, and very coarsely crosshatched appeared with both the Netherlands and the Illinois isolations in platings from pure cultures. This form was very stable and has never been observed to revert to either of the other forms. It was very weakly pathogenic.

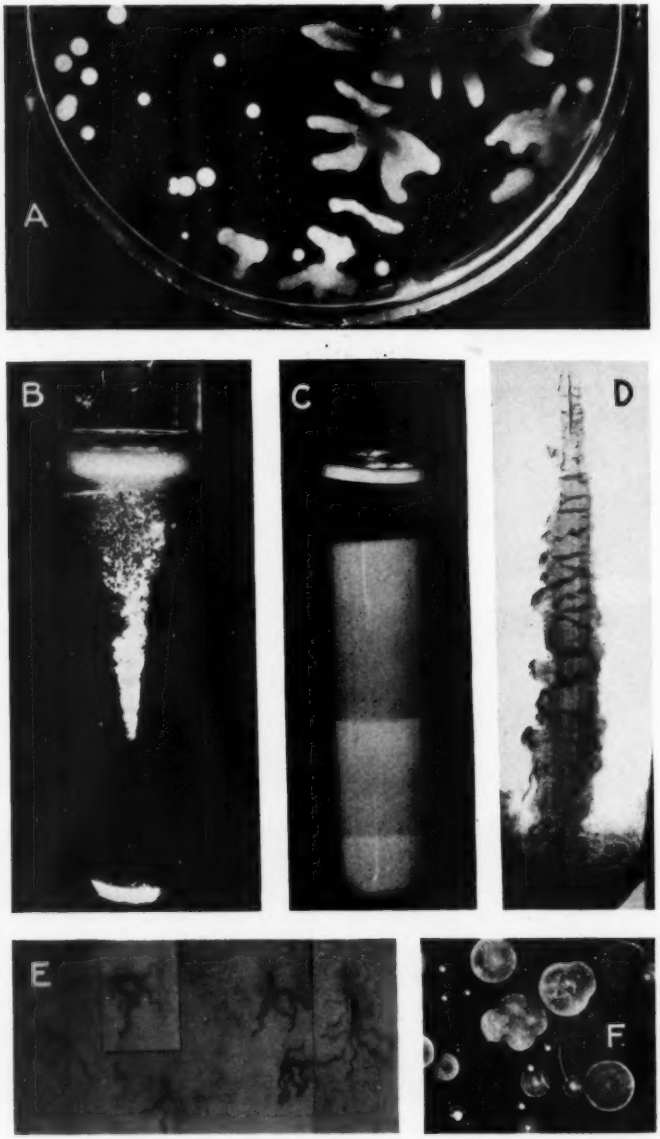
BEEF-AGAR SLANTS.—The same difference in surface in the two strains was apparent on beef-agar slants as on plates. (Pl. 2, D.) Growth is 3 to 7 mm. wide, white, transparent, opalescent, finely crosshatched by oblique light, with wavy or scalloped margins. The whole of the agar becomes green. Small crystals form in abundance just below the growth on the slant and below the V.

BEEF-AGAR STABS.—Growth characteristic of each strain occurs on the surface, finally covering it. In the stab, scanty growth is visible for less than one-third the length of the needle track. Small crystals form just below the surface of the agar.



Colonies of *Bacterium syringae* on beef-infusion agar

A. Fimbriate colony, 4 days old, natural size
B. Illinois strain, 24 hours old. Colonies of the Holland strain are indistinguishable from these at this age
C. Illinois strain, 2 days old
D. Holland strain 2 days old
E. Illinois strain, 3 days old
F. Holland strain, 3 days old
G. Holland strain, 5 days old
H. Illinois strain, 5 days old
(B to H, × 10)



A. Round and fimbriate colonies on beef-infusion agar plate, 2 days old
B. Gelatin stab, 48 hours old at 18° C.
C. Litmus milk culture, 4 days old
D. Beef-infusion agar slant, 3 days old
E. *Bacterium syringae* showing polar flagella
F. Colonies on beef-infusion gelatin plate, 2 days old

BEEF BROTH.—In peptone beef-infusion broth clouding is evident within 24 hours. Green fluorescence begins at the top on the second day, and a delicate filmy shining wrinkled pellicle forms which breaks into fragments readily on shaking. This pellicle is characteristic of wrinkled colonies only. Other types of colony have a dull white opaque pellicle which disappears as a fine cloud on shaking. All types produce the green fluorescence. Numerous small crystals lie in the white precipitate.

BEEF-EXTRACT MEDIA.—Growth is less vigorous on beef-extract agar than on beef-infusion agar, colonies attaining only approximately one-half the size under the same conditions for growth. No surface wrinkling occurs and no green fluorescence develops on plates, slants, or in broth made with beef extract, and only occasional small crystals are formed.

POTATO CYLINDERS.—On steamed potato cylinders growth is thin, grayish white, never becoming abundant. The potato is browned.

COHN'S SOLUTION.—There is very weak growth in Cohn's solution, evidenced by a faint but decided clouding and a very small quantity of white precipitate.

USCHINSKY'S SOLUTION.—Clouding is moderate to heavy in this medium, with a delicate rim and pellicle and a very pronounced green color. An abundant white precipitate is formed. No crystals were observed. The Netherlands strain gave heavier growth than the Illinois strain, as evidenced by both clouding and pellicle.

FERMI'S SOLUTION.—In Fermi's solution a heavy pellicle is formed, but the fluid is only moderately clouded and takes on a beautiful blue-green fluorescence.

BLOOD SERUM.—There is moderate growth on blood serum, but no liquefaction takes place.

PHYSIOLOGICAL CHARACTERS

LIQUEFACTION OF GELATIN.—In beef-peptone-gelatin stabs, made with both beef extract and beef infusion, liquefaction begins within 24 hours at 18° C. and is infundibuliform. Within a day or two it becomes stratiform at the top with a narrow funnel of liquefaction reaching one-half the depth of the gelatin or more and largely filled with growth. (Pl. 2, B.) When inoculations are made from freshly isolated cultures, liquefaction is rapid, often complete in 6 days, but when old isolations are used liquefaction progresses much more slowly and occasionally never is really complete, although the start is prompt and infundibuliform. The surface layer is green fluorescent. There is complete liquefaction and green fluorescence throughout in 6 to 16 days. With cultures which had been out of the plant for months, liquefaction began promptly at 18°, but progressed much more slowly and sometimes was not complete after six weeks.

FERMENTATION OF SUGARS.—Fermentation was tested on beef-extract agar and synthetic (ammonium phosphate) agar containing brom cresol purple with saccharose, dextrose, maltose, lactose, galactose, levulose, glycerin, and mannit. Acid was produced, beginning on the second day, from all except lactose and maltose, the agar becoming bright yellow throughout. Glycerin and mannit were somewhat less acid than the others. Cultures were held for two weeks. In the beef-extract agar, but not in the synthetic agar, alkali production began in five days and extended through two-thirds of the culture in saccharose, dextrose, galactose, and levulose. The two strains were alike with all the sugars tested.

In fermentation tubes containing 1 per cent peptone water with the same sugars there was no gas. Clouding appeared in the closed end with saccharose and dextrose only, heavier in saccharose in both strains.

RELATION TO FREE OXYGEN.—The organism is not strictly aerobic, since growth takes place in the closed end of fermentation tubes with saccharose and dextrose. In stab cultures, however, there is only scanty growth about one-third the depth of the needle track even when the sugars mentioned above are present.

HYDROLYSIS OF STARCH.—There is very slight diastatic action. Starch-agar plates seven days old, when flooded with iodine solution, show a purple area 3 to 8 mm. wide around the growth, while on potato cylinders growth is scanty.

NITRATE REDUCTION.—There is no nitrite reaction when 10-day-old nitrate beef-broth cultures are tested with the starch-potassium iodide-sulphuric acid method, but a very decided though very moderate nitrite reaction when the α-naphthalymine-sulphanilic acid test was used. This would indicate very weak nitrate reduction.

ACID IN MILK.—No acid is formed in milk, but a slight alkalinity is evident in litmus milk.

REDUCTION OF LITMUS.—Litmus milk is blued slightly. A layer of clear whey 3 to 5 mm. forms at the top, but there is no further evidence of coagulation. Peptonization then begins and progresses downward in bands (pl. 2, C), followed by reduction of the litmus. Peptonization and reduction are complete within two weeks. Within the next two weeks the color returns to a dark grayish blue and remains so indefinitely.

REDUCTION OF METHYLENE BLUE.—Methylene blue in milk is reduced promptly and completely, the reduction beginning by the second day and becoming complete on the third or fourth day. By the fourth week the color returns a clear green (forest green according to Ridgway (*4*)).

AMMONIA PRODUCTION.—Ammonia production is strong in beef-peptone media.

HYDROGEN SULPHIDE.—Hydrogen sulphide is produced very scantily or not at all.

PRODUCTION OF INDOL.—No indol was produced in Dunham's solution within 20 days, tests being made at intervals of 5 days.

TOLERATION OF ACID.—Tests were made in beef-infusion broth and in beef-extract broth with a series of P_H values. Growth occurred in beef-infusion broth from P_H 5.4 to P_H 9.1, with the optimum at P_H 6.8, and in extract broth from P_H 5.3 to 9, with the optimum at P_H 6.5 to 7.

TOLERATION OF SODIUM CHLORIDE.—There is clouding within 24 hours in beef broth containing 1, 2, and 3 per cent NaCl, in 2 days in 4 per cent, and after 5 days in 5 per cent. Very slight growth takes place in 6 per cent. In 3 to 6 per cent NaCl growth consists of distorted irregularly swollen filaments.

RESISTANCE TO DESICCATION.—The organism is very sensitive to drying. Drops of a 24-hour-old culture placed on sterile cover glasses in sterile Petri dishes and kept in the dark gave no clouding when dropped into beef broth after 2 days' drying.

TEMPERATURE RELATIONS.—Best growth occurs at 28° to 30° C. Beef-broth cultures kept at 35°, 36°, and 37° for 8 days remained clear. When removed to room temperature (28° C.) the 35° cultures clouded but the 36° and 37° cultures remained clear. Within 24 hours clouding was visible in beef-broth cultures at temperatures from 10° to 30° and after 5 days at 1° to 2.5°. The thermal death point is 51° for both strains.

BRIEF CHARACTERIZATION OF BACTERIUM SYRINGAE [1]

Bacterium syringae is a motile rod 1.2 to 1.8 by 0.6 μ with one to several polar flagella; single, in pairs or short chains. Long twisted filaments and chains occur in salt bouillon. No spores are formed. A small capsule may be demonstrated by Ribbert's dahlia stain. The organism is Gram-negative, but stains readily with the ordinary bacterial stains. An irregular or polar staining is obtained with carbol fuchsine. Agar colonies round or fimbriate, smooth or wrinkled, white; liquefies gelatin rapidly; produces green fluorescence in beef-infusion media; reduces litmus and methylene blue in milk; forms a soft rennet curd in milk and clears it in bands; produces ammonia but no indol or hydrogen sulphide; weak nitrate reduction; feeble diastatic action on starch; grows well in Uschinsky's solution and Fermi's solution, but very feebly in Cohn's solution; produces acid from saccharose, dextrose, galactose, levulose, glycerin, and mannit, but not from lactose or maltose within two weeks; no gas; tolerates 5 per cent NaCl in beef broth; optimum temperature 28° to 30° C., maximum 35°, minimum below 1°.

SUMMARY

A lilac blight occurring in Illinois has been proved to be identical with the lilac blight of Europe caused by *Bacterium syringae* (Van Hall) EFS.

The disease blackens and kills young stems and leaves in the spring. It is most destructive in rainy seasons. The appearance of affected plants suggests fire blight.

The blight is primarily a disease of the parenchyma, but it also invades the vascular tissue.

Careful pruning out of diseased twigs is the best method of control.

Cultural characters of the organism are given.

[1] "Bacterium" is here used according to the system of nomenclature proposed by Erwin F. Smith (*8, p. 171*) and used by him for this organism in the same volume (*8, p. 63*).

Bacterium citriputeale has been found indistinguishable from *Bact. syringae* in cultures, and cross inoculations with the two organisms have produced good infections.

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FURTHER EXPERIMENTS WITH SEED TREATMENTS FOR SWEET-CORN DISEASES[1]

By C. S. REDDY, *Associate Pathologist*, and J. R. HOLBERT, *Agronomist, Office of Cereal Crops and Diseases, Bureau of Plant Industry, United States Department of Agriculture*[2]

INTRODUCTION

This paper presents the results from additional experiments with seed treatments for the control of certain seed-borne diseases of sweet corn (*Zea mays saccharata*). These experiments were conducted in 1926 in the greenhouse and field at the Arlington Experiment Farm, Rossyln, Va., and in the field near Bloomington, Ill.[3]

A previous paper[4] on this subject dealt for the most part with experiments conducted in 1925. For several reasons, it was thought that the 1925 results from seed treatments for sweet-corn diseases were not indicative of the full possibilities or even the average possibilities of such treatments for increasing yields from diseased seed. The spring of 1925 was exceptionally dry, and the plantings were made late when the soil had warmed to a considerable degree. In moderately dry warm soil both *Diplodia zeae* and *Gibberella saubinetii* produce minimum effects as seedling blights of corn.[5] [6]

The 1926 results, presented in this paper, were obtained for the most part under conditions favorable to the development of Diplodia seedling blight. For this reason, the addition of these results to those already published should give a basis for a more accurate estimate of the value of seed treatments for the control of certain sweet-corn diseases.

PRELIMINARY SEED-TREATMENT EXPERIMENTS IN GREENHOUSE

One of the early difficulties encountered in experimenting with seed treatments for corn was to find a satisfactory means of limiting the number of seed-treatment materials to be used in field experiments without omitting possibly the best ones.

Short-time preliminary experiments were conducted in the greenhouse for this purpose. Cylindrical cans 8 inches in diameter and 10 inches in height were filled with moist soil to within 4 inches of the top. A wire cage or basket of ½-inch mesh was set on the soil and 2 inches of wet sand placed in this. Twenty-five kernels of the corn under experimentation were laid on this sand and then covered with another inch of wet sand. (Fig. 1.) Four cans were used for each treatment and more than four for each control.

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[1] Received for publication Nov. 29, 1927; issued April, 1928.

[2] The writers wish to express their indebtedness to A. G. Johnson, of the Office of Cereal Crops and Diseases, Bureau of Plant Industry, United States Department of Agriculture, for advice, helpful suggestions, and assistance in preparing the manuscript.

[3] The investigations conducted near Bloomington, Ill., were in cooperation with Funk Brothers Seed Co.

[4] REDDY, C. S., HOLBERT, J. R., and ERWIN, A. T. SEED TREATMENTS FOR SWEET-CORN DISEASES. Jour. Agr. Research 33: 769–779, illus. 1926.

[5] HOLBERT, J. R., BURLISON, W. L., KOEHLER, B., WOODWORTH, C. M., and DUNGAN, G. H. CORN ROOT, STALK, AND EAR ROT DISEASES, AND THEIR CONTROL THRU SEED SELECTION AND BREEDING. Ill. Agr. Expt. Sta. Bul. 255, p. 239–478, illus. 1924.

[6] DICKSON, J. G. INFLUENCE OF SOIL TEMPERATURE AND MOISTURE ON THE DEVELOPMENT OF THE SEEDLING-BLIGHT OF WHEAT AND CORN CAUSED BY GIBBERELLA SAUBINETII. Jour. Agr. Research 23: 837–870, illus. 1923.

Usually it was not necessary to add water during the period of the experiment, which was about 16 days. When necessary, an equal quantity of water was added to each can. The seed used was Country Gentleman sweet corn, which, on the germinator, had shown 88 per cent viability and 50 per cent infection with *Diplodia zeae*. Wire cages of somewhat smaller mesh might have been better than those used. In similar experiments with dent corn, conducted by the junior writer, the procedure was varied from that described above by adding the sand in a dry condition, moistening with measured quantities of water, and then adding a light sprinkling of dry sand to serve as a mulch.

When Diplodia-infected seed is used, the most satisfactory results are obtained if greenhouse temperatures are between 22 and 25° C. and the moistures are kept at 60 to 65 per cent of the water-holding capacities of sand and soil. These conditions insure maximum germination of Diplodia-infected seed, good early growth, and the greatest development of Diplodia seedling blight.[7] Moderately high soil moisture is very important, but it should not exceed 67 to 70 per cent of the water-holding capacity.

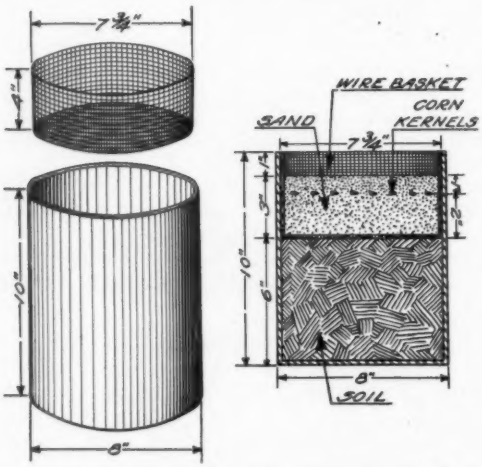

FIG. 1.—Soil can, wire basket, and diagram showing their use in preliminary seed-treatment experiments

When Gibberella-infected seed is used, the greenhouse temperature should be maintained at 15 to 17° C., with the soil moisture about the same as or somewhat lower than for the Diplodia-infected seed.

The seed-treatment materials and methods used were:

1. Uspulun, 0.5 per cent, one and one-half hour soak.
2. Semesan, 0.5 per cent, one and one-half hour soak.
3. Mercury A, 0.5 per cent, one and one-half hour soak.
4. Bayer, dust.
5. Semesan, dust.
6. Semesan Jr., dust.
7. Semesan 13 UG, dust.
8. Semesan 13 UH, dust.
9. Du Pont 12, dust.
10. Du Pont 12 Bel–10 (Semesan Bel), dust.
11. Du Pont 12 Bel–6, dust.
12. Du Pont 12 Bel A, dust.
13. Du Pont 12 Bel B, dust.
14. Du Pont 35, dust (included only for field experiments).
15. Du Pont 50, dust.
16. Du Pont 51, dust.
17. Du Pont 57, dust.
18. Du Pont 62, dust.
19. Du Pont 63, dust.
20. Du Pont Z4, dust.
21. Mercury C, dust.
22. Corona 640, dust.
23. Abavit B, dust.
24. S. F. A. 225, dust.
25. S. F. A. 225 V, dust.
26. S. F. A. AZ III, dust.
27. S. I. 220, dust.

[7] HOLBERT, J. R., KOEHLER, B., and DUNGAN, G. H. THE DIPLODIA DISEASE OF CORN. Ill. Agr. Expt. Sta. (Unpublished manuscript.)

Nos. 1 and 4 were obtained from the Bayer Co. (Inc.), New York, N. Y.; 2 and 5 to 20, inclusive, from E. I. du Pont de Nemours & Co. (Inc.), Wilmington, Del.; 3 and 21 from Roessler & Hasslacher Chemical Co., Perth Amboy, N. J.; 22 from Corona Chemical Division, Pittsburgh Plate Glass Co., Milwaukee, Wis.; 23 from Chemische Fabrik Ludwig Meyer, Mainz, Germany; 24 to 26, inclusive, from Saccharin-Fabrik, Actiengesellschaft, Magdeburg, Südost, Germany; and 27 from Actiengesellschaft für Anilin Fabrikation, Wolfen, Kreis Bitterfeld, Germany.

When the treatment controlled the disease without injury to the seed, the roots grew rapidly through the sand into the soil and an

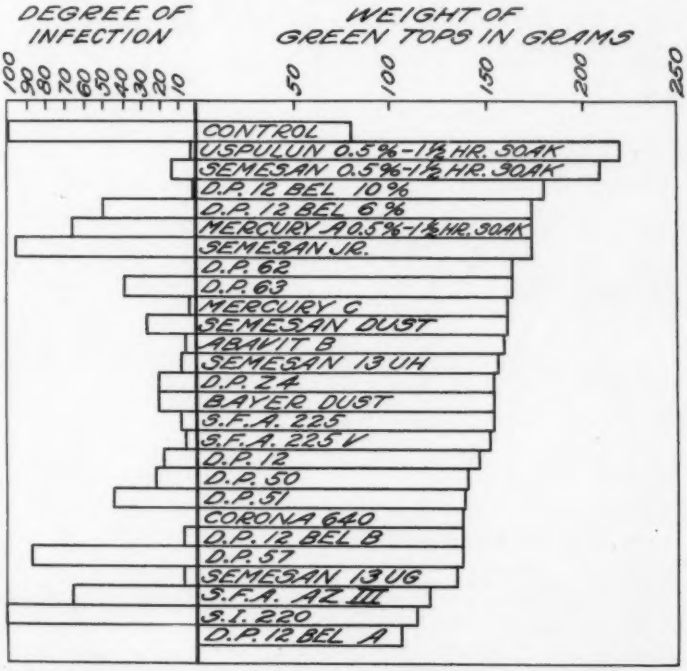

FIG. 2.—Results of preliminary seed-treatment experiments with Diplodia-infected Country Gentleman sweet-corn seed, showing the weights of green tops and the degrees of infection in proportion to those of the control (taken as 100)

early development of the aerial parts resulted. When no treatment was applied or when the treatment did not control the disease, the infected plants grew slowly. Often the roots did not reach the soil, and as a result the growth of the aerial portions was retarded. Sometimes the treatment controlled the disease but exerted a depressing effect on the growth of the plants. In this case also the top growth was retarded.

At the end of about 16 days the weights of green tops were obtained, the wire cages were removed, and the sand was washed out by a stream of water from a hose so that the parts of the plants below the surface of the sand were exposed for observation. Data then were

taken on the number and the severity of the infections. At this stage most of the lesions are on the mesocotyls.

The data from these preliminary experiments are presented graphically in Figure 2. In this graph the results, except those from the control series, are arranged in a graduated series according to the weight of green tops obtained. The degree of infection is relative to the infection found in the controls, which is arbitrarily taken as 100. The plants are shown in Figure 3.

In these preliminary tests the best treatment materials are indicated by high weights of tops and low degrees of infection. In general, the results of these preliminary experiments indicated that

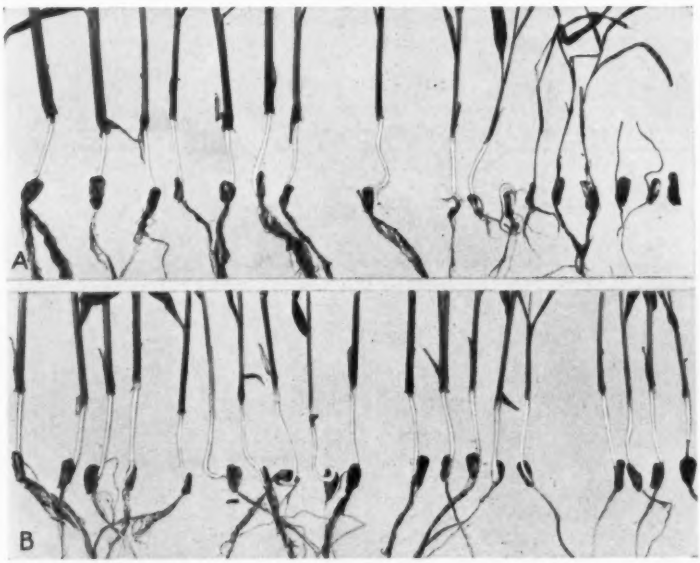

FIG. 3.—Country Gentleman sweet-corn plants, 16 days old, grown in the greenhouse from Diplodia-infected seed, using the method and apparatus illustrated in Figure 1, at Arlington Experiment Farm, Rosslyn, Va., January, 1927: A—Plants from untreated seed, showing all the plants from one of the control cans. Of the 25 kernels planted, 17 produced plants. Of these, 7 were uninfected, 1 slightly infected, 3 moderately infected, and 6 severely infected (dead). B—Plants from seed treated with organic-mercury dust showing all the plants from one can. Of the 25 kernels planted, 18 produced plants. Of these, 12 were uninfected, 5 slightly infected, 1 moderately infected, none severely infected (dead)

Uspulun and Semesan were practically equally efficient as liquid disinfectants. A number of dusts gave promising indications of control. However, some of the dusts seemed to show depressing effects on the growth of tops, or did not control infections, or showed both reactions.

The results from the last seed treatment (Du Pont 12 Bel A) represented in Figure 2 show that the disease was controlled, but that the weight of tops produced was lowest. This low weight of tops probably is the result of an injurious effect of the treatment. The treatment that resulted in the next lowest weight of tops permitted a high degree of infection. It is likely in this case that lack of disease control rather than an injurious effect on the plants caused the small development of tops.

TIME-OF-PLANTING AND SEED-TREATMENT EXPERIMENT IN FIELD

MATERIALS AND METHODS

A combined time-of-planting and seed-treatment experiment was conducted in the field at the Arlington Experiment Farm, Rosslyn, Va., in which specially selected Diplodia-infected Country Gentleman seed grown at Bloomington, Ill., in 1925 was used. Plantings were made on April 6, 14, and 22, and May 7, 1926. Half of the seed was dusted with S. F. A. 225 at the rate of 2 ounces per bushel. The treatment was made by shaking the seed and the dust in a closed container for 10 minutes. Each planting consisted of 12 rows of 19 hills each, the hills being spaced 40 inches apart each way. Six rows were planted with treated seed, and untreated seed was used in alternate rows, making six replications in each of the four plantings. Three kernels were planted to the hill. Stand data were taken when the plants were about 6 inches high. Yield data were taken when the corn was prime for canning. Total yields were obtained by weighing the snapped ears without removing the husks. Yields of corn prime for canning were obtained after the husks were removed. These latter yields did not include small nubbins, rotten ears, and ears which had not reached or had passed the prime stage.

SOIL MOISTURE AND TEMPERATURE RELATIONS

The daily maximum and minimum soil temperatures at a depth of 1 inch and the daily rainfall for the months of April and May are given in Table 1.

TABLE 1.—*Daily maximum and minimum soil temperatures at a depth of 1 inch and daily rainfall for April and May, 1926, at Arlington Experiment Farm, Rosslyn, Va.*

Date	Soil temperatures (° C.)		Rainfall (inches)	Date	Soil temperatures (° C.)		Rainfall (inches)
	Maximum	Minimum			Maximum	Minimum	
Apr. 1			0.07	May 1	30	11	
Apr. 2				May 2	30	13	
Apr. 3				May 3	27	7	
Apr. 4			.02	May 4	24	3	
Apr. 5				May 5	27	7	
Apr. 6	15	2		May 6	27	11	
Apr. 7	17	9	.16	May 7	34	12	
Apr. 8	18	7	.17	May 8	33	12	
Apr. 9	17	2	.27	May 9	30	13	
Apr. 10	20	7		May 10	23	9	
Apr. 11	12	1		May 11	26	7	
Apr. 12	13	2	.17	May 12	28	13	
Apr. 13	8	3		May 13	30	9	
Apr. 14	18	4	.02	May 14	28	11	0.13
Apr. 15	15	0		May 15	30	12	2.41
Apr. 16	20	5		May 16	15	7	
Apr. 17	19	5		May 17	25	11	
Apr. 18	9	0		May 18	27	14	
Apr. 19	11	−1	.05	May 19	26	12	.15
Apr. 20	14	2		May 20	21	9	
Apr. 21	24	6		May 21	22	9	
Apr. 22	28	10		May 22	26	10	
Apr. 23	24	10		May 23	17	6	.01
Apr. 24	28	9		May 24	22	16	
Apr. 25	23	6	.01	May 25	30		
Apr. 26	21	3		May 26		8	
Apr. 27	24	9		May 27	25	12	
Apr. 28	15	3		May 28	26	12	
Apr. 29	21	4	.03	May 29	24	7	
Apr. 30	25	11		May 30	26	8	
				May 31	26	14	

Table 1 shows that the soil was cold for a number of days following the first planting, April 6. As a result, the first and second plantings, made April 6 and 14, emerged at the same time. It is of interest to note later the effect of these severe conditions at planting time on the field stands and yields from untreated and treated seed. In general, the early growth of the first series (April 6) was in cold, moderately moist soil; that of the second (April 14) and third (April 22) series

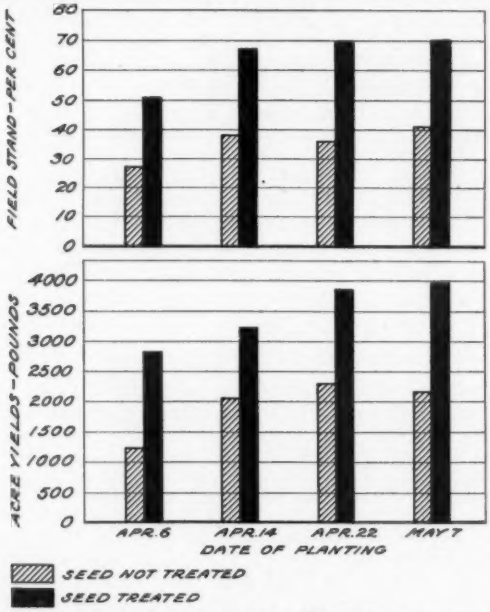

FIG. 4.—Field stands and acre yields of Country Gentleman sweet corn, prime for canning, with husks removed, grown from Diplodia-infected seed, untreated or treated with S. F. A. 225 dust and planted on four different dates at Arlington Experiment Farm, Rosslyn, Va., in 1926. (Data from Tables 2 and 3)

was in moderately cold, dry soil; and that of the fourth series (May 7) was in moderately warm, wet soil. If any of the series had been planted in warm, moderately dry soil, very little Diplodia seedling blight would have been expected to develop and, as a result, there would have been very little increase in the yields from treated seed. However, as these two conditions, namely, warm and moderately dry soil, did not obtain simultaneously during the critical periods of these experiments, it would be expected that Diplodia seedling blight would be active in each series and that significant increases in yield would be obtained from the treated seed, if the treatment controlled the disease without injuring the seed.

DATA ON STAND AND YIELD

The stand data from this seed-treatment experiment, which involves four series of plantings of untreated and treated Diplodia-infected Country Gentleman seed, are presented in Table 2 and Figure 4. The yield data are presented in Table 3 and shown graphically in Figure 4.

TABLE 2.—*Mean field stands from Diplodia-infected Country Gentleman seed corn, untreated or treated with S. F. A. 225 dust and planted at Arlington Experiment Farm, Rosslyn, Va., on four different dates in 1926*

Date of planting	Soil conditions following planting	Field stand average of 6 replications (per cent)		
		Untreated seed	Treated seed	Increase due to seed treatment
Apr. 6	Cold and moderately moist	27.2	51.2	88.2
Apr. 14	Cold and moderately dry	38.0	67.9	78.7
Apr. 22	do	36.8	69.3	88.3
May 7	Warm and moderately wet	41.5	70.8	70.6

TABLE 3.—*Acre and plot yields of snapped ears and of husked ears prime for canning, grown from Diplodia-infected Country Gentleman seed corn, untreated or treated with S. F. A. 225 dust, as planted in six replications in 1926, at Arlington Experiment Farm, Rosslyn, Va.*

Data compared	Untreated seed		Treated seed	
	Total ears, husks not removed	Ears prime for canning, husks removed	Total ears, husks not removed	Ears prime for canning, husks removed
Series 1, planted Apr. 6:				
Plot yields, 6 replications, pounds	15.8	5.1	29.5	13.0
	17.8	4.8	32.4	14.1
	15.8	6.8	37.8	12.6
	16.8	6.5	41.2	15.8
	15.3	6.5	41.0	13.0
	17.2	6.2	36.5	12.8
Total, do	98.7	35.9	235.4	81.3
Mean acre yields, do	3,463	1,260	7,909	2,853
Net increase, do			4,446	1,593
Increase, per cent			128.4	126.4
Odds			9,999:1	9,999:1
Series 2, planted Apr. 14:				
Plot yields, 6 replications, pounds	30.1	11.0	41.6	17.5
	25.0	9.0	48.3	19.6
	22.0	9.0	35.1	13.1
	24.2	9.0	36.0	16.0
	29.2	11.5	32.5	12.9
	24.0	8.8	33.5	13.2
Total, do	154.5	58.3	227.0	92.3
Mean acre yields, do	5,421	2,046	7,965	3,239
Net increase, do			2,544	1,193
Increase, per cent			46.9	58.3
Odds			336:1	295:1
Series 3, planted Apr. 22:				
Plot yields, 6 replications, pounds	24.5	12.2	32.5	18.2
	23.5	12.5	35.6	19.5
	22.5	13.1	31.3	16.8
	26.5	12.8	35.7	19.8
	13.7	6.0	26.7	14.9
	16.2	9.5	40.5	21.0
Total, do	126.9	66.1	202.3	110.2
Mean acre yields, do	4,453	2,319	7,099	3,867
Net increase, do			2,646	1,548
Increase, per cent			59.4	66.8
Odds			521:1	1,683:1
Series 4, planted May 7:				
Plot yields, 6 replications, pounds	19.6	8.6	39.5	19.8
	24.8	11.5	46.1	22.0
	26.8	11.5	38.0	18.1
	24.5	10.9	39.0	20.5
	19.0	9.0	29.8	13.0
	23.0	10.5	42.6	20.0
Total, do	137.7	62.0	235.0	113.4
Mean acre yields, do	4,832	2,176	8,246	3,979
Net increase, do			3,414	1,803
Increase, per cent			70.7	82.9
Odds			4,201:1	3,332:1

TABLE 3.—*Acre and plot yields of snapped ears and of husked ears prime for canning, grown from Diplodia-infected Country Gentleman seed corn, untreated or treated with S. F. A. 225 dust, as planted in six replications in 1926, at Arlington Experiment Farm, Rosslyn, Va.*—Continued

SUMMARY OF YIELD DATA

Series	Date of planting, 1926	Acre yield of prime corn (pounds)		Increase from treatment		Odds
		Untreated seed	Treated seed	Pounds per acre	Per cent	
No. 1	Apr. 6	1,260	2,853	1,593	126.4	9,999:1
No. 2	Apr. 14	2,046	3,239	1,193	58.3	295:1
No. 3	Apr. 22	2,319	3,867	1,548	66.8	1,683:1
No. 4	May 7	2,176	3,979	1,803	82.9	3,332:1
Mean		1,950.25	3,484.5	1,534.25	78.7	

Tables 2 and 3 and Figure 4 show that stands and yields were significantly increased as a result of dusting the seed with S. F. A. 225. Although the first planting was made under such unfavorable conditions that the stands and yields from untreated seed were very low, the total yield from the treated seed was practically the same as the total yield from treated seed in the more favored plantings. The yield of prime canning corn from treated seed in the first planting is higher than the yields of prime canning corn from untreated seed in any of the plantings. These data indicate that the practice of treating the seed may make feasible the practice of earlier planting of sweet corn, thereby solving certain farm-management problems and sometimes extending the canning season.

EXPERIMENTS WITH COMMERCIAL SWEET-CORN SEED

Fifteen pounds each of two bulk lots of shelled seed of Country Gentleman were bought on the market in Washington, D. C. Both lots cost the same. By germinating 200 kernels from each, it was determined that lot 1 germinated 100 per cent and was nearly disease free, and that lot 2 germinated 90 per cent and 20 per cent of the kernels were Diplodia infected.

A seed-treatment experiment with these two lots of seed was conducted in the field near Bloomington, Ill. The 9 treatment materials were as follows: S. F. A. 225 dust, Semesan 13 UG dust, Semesan Jr. dust, Bayer dust, Du Pont 12 Bel dust, Abavit B dust, Du Pont 35 dust, Semesan, 0.5 per cent for a one and one-half hour soak, and Uspulun, 0.5 per cent for a one and one-half hour soak. The dusts were applied at the rate of 2 ounces per bushel and shaken with the seed in a closed container for 10 minutes. The plots were planted in rows of 22 hills each at the rate of 3 kernels per hill in hills spaced 42 inches each way. Since there were 6 replications of each treatment and 12 replications without treatment, 132 plots were required for the experiment. The yield data were obtained by the same methods used in obtaining the data presented in Table 3. The yields of prime canning corn are given in Table 4 and presented graphically in Figure 5.

TABLE 4.—*Mean acre yields (average of six replications) of Country Gentleman sweet corn, prime for canning, with husks removed, grown from two lots of commercial seed, untreated or treated with each of several seed-treatment materials, respectively, as planted near Bloomington, Ill., in 1926*

Seed treatment	Acre yield of prime corn with husks removed (pounds)		Increase in acre yield following treatment		Odds
	Untreated seed	Treated seed	Pounds	Per cent	
Lot 1, better seed					
Dusts:					
Semesan Jr.	1,972.5	2,344.4	371.9	18.9	11:1
Bayer dust	2,344.4	2,473.7	129.3	5.5	3:1
Du Pont 12 Bel.	2,344.4	2,392.9	48.5	2.1	2:1
Du Pont 35	2,215.0	2,328.2	113.2	5.1	3:1
S. F. A. 225	1,972.4	2,231.1	258.7	13.1	13:1
Semesan 13 UG	1,972.4	2,279.6	307.2	15.6	19:1
Abavit B	2,215.0	2,312.0	97.0	4.4	2:1
Soaks:					
Semesan	1,972.5	2,150.3	177.8	9.0	5:1
Uspulun	1,972.5	2,409.0	436.5	22.1	28:1
Mean	2,109.0	2,324.6	215.6	10.2	
Lot 2, poorer seed					
Dusts:					
Semesan Jr.	1,390.4	2,037.1	646.7	46.5	1,193:1
Bayer dust	1,293.4	1,616.8	323.4	25.0	21:1
Du Pont 12 Bel.	1,293.4	1,681.4	388.0	30.0	55:1
Du Pont 35	1,293.4	1,600.6	307.2	23.8	14:1
S. F. A. 225	1,390.4	1,681.4	291.0	20.9	62:1
Semesan 13 UG	1,390.4	1,923.9	533.5	38.4	339:1
Abavit B	1,293.4	1,665.3	371.9	28.8	18:1
Soaks:					
Semesan	1,196.4	1,746.1	549.7	45.9	28:1
Uspulun	1,196.4	1,455.1	258.7	21.6	6:1
Mean	1,304.2	1,712.0	407.8	31.3	

As shown in Table 4 and Figure 5, certain seed treatments caused significant increases in yield. However, they were decidedly more beneficial to the poor commercial seed (lot 2) than to the good commercial seed (lot 1). The dust treatments were more consistent in their beneficial results than the liquid treatments, as judged by the odds obtained.

DISCUSSION

The results of experiments herein reported show that Diplodia seedling blight of sweet corn can be largely prevented by dust or liquid treatment of the seed with any one of several compounds, without injury even to the best seed. Conditions unfavorable for plant growth at planting time seem to increase the relative benefits from seed treatments. There are indications that dust-treated sweet-corn seed can remain longer in cold soils without injury than untreated seed. Hence, satisfactory stands may be obtained from dust-treated seed in earlier plantings than can be obtained from untreated seed. Dust seed treatments for the control of Diplodia seedling blight of sweet corn are as satisfactory as liquid treatments and possess certain advantages. They are easily applied and obviate the risk of injury to germination involved in wetting and drying the seed. Dust treatments can be made long in advance of planting time without injury to the seed, and tend to prevent insect infestation while such treated seed is stored. These features of the dust

method especially recommend it for use at central treating plants and by commercial firms handling the seed in quantity.

With sweet corn, as with dent corn, it is highly desirable to determine to what extent seed stock is infected with Diplodia and Gibberella, and to separate out the nearly disease-free seed in a manner now extensively used for dent corn. As with dent corn, nearly disease-free seed is not benefited by seed treatments, while second-grade seed usually is greatly benefited by the proper seed treatments.

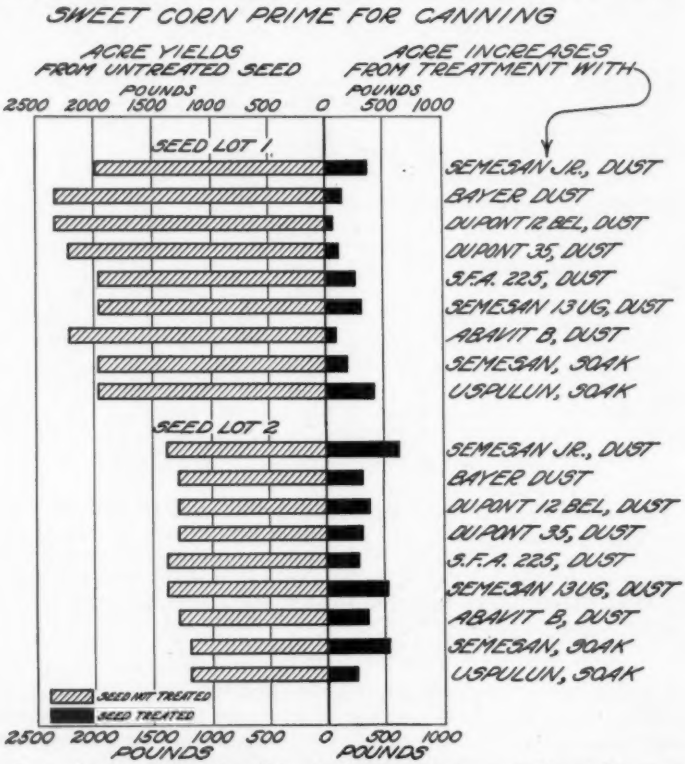

FIG. 5.—Acre yields of husked ears of Country Gentleman sweet corn, prime for canning, grown from treated and untreated seed of two commercial lots. (Data from Table 4)

The diseases caused by Diplodia and Gibberella are so important on sweet-corn seed that some commercial canners follow the practice of making ear germination tests of their sweet-corn seed stock and, on the basis of such tests, separate their seed into three lots, as follows:

No. 1. Nearly disease-free seed.

No. 2. Seed that germinates well, but carries some infection with Diplodia, Gibberella, and other similar organisms.

No. 3. Badly diseased seed.

The No. 1 seed is planted without seed treatment, the No. 2 seed is treated before planting, and the No. 3 seed is discarded or marketed, if any market can be found for it.

SUMMARY

Satisfactory preliminary experiments to determine the relative merits of seed-treatment materials can be conducted in a greenhouse, if seed of sweet corn of good germination, highly infected with *Diplodia zeae*, is available.

Yields of prime canning corn from Diplodia-infected Country Gentleman seed treated with an organic-mercury dust were 126.4 per cent greater than yields from similar untreated seed when the planting was made in cold, moderately moist soil; 58.3 and 66.8 per cent greater when plantings were made in moderately cold, dry soil; and 82.9 per cent greater when plantings were made in warm, moderately wet soil.

Of two commercial lots of Country Gentleman seed, treatments were decidedly more beneficial to the corn grown from the poorer seed lot. Following treatment of the seed, the average increase in yield from the good lot was 10.2 per cent and from the poor lot 31.3 per cent over that from the untreated seed.

Dust treatments are more consistent in their beneficial effects, are more easily applied, and involve less risk of injury to the seed than liquid treatments.

THE APPLE CURCULIO AND ITS CONTROL BY HOGS[1]

By B. B. FULTON, *Iowa State College*

INTRODUCTION

The importance of the apple curculio (*Tachypterus quadrigibbus* Say) has usually been overshadowed by the emphasis given to the plum curculio (*Conotrachelus nenuphar* Herbst). While the latter does not thrive well under the conditions of modern orchard practice, the former seems to be affected very little if at all by spraying, soil cultivation, and pruning.

The destructiveness of the apple curculio in Iowa was first called to the writer's attention by R. M. Clark, manager of the Apple Grove Orchards, south of Mitchellville, Iowa. The plum curculio had been reduced to negligible numbers in this orchard but the manager estimated the loss caused by the apple curculio as at least a thousand dollars a year. This loss was due in part to the added cost of sorting out the unmarketable injured fruit.

The purpose of this paper is to report new observations on the seasonal history and habits of the apple curculio and to describe an effective method of control which can be easily applied over much of the territory in which this insect has proved to be destructive. A description of the stages of its development and many other previously reported facts concerning its life history have been omitted.

REVIEW OF LITERATURE

The first important publication on the apple curculio was by Riley (*7*).[2] He describes and figures the main life stages and describes the punctures made in the green fruit, and the scars on mature pears identified as the early work of the apple curculio. It is evident, however, that his information on the life history is not derived from observations in the orchard. He states that infested apples never fall and that pupation takes place in the apple while it is still on the tree. Gillette (*5*) gives an account of the insect's work on apple and describes its oviposition habits. Crandall (*4*) has made the most complete study of the life history of the apple curculio in connection with his work on the plum curculio. In one respect his observations do not agree with those recorded here. He states that his new generation of beetles fed on the fruit to a very limited extent. Moreover, he describes and figures a type of injury which he ascribes entirely to the plum curculio, but which, in the opinion of the present writer, is more typical of that caused by the late summer feeding of the apple curculio.

The apple curculio has been mentioned as a pest of the apple in a number of widely separated localities. Its injury seems to be most severe in Missouri, Kansas, Illinois, Iowa, Maine, and the Province

[1] Received for publication Dec. 2, 1927; issued April, 1928.

[2] Reference is made by number (italic) to "Literature cited," p. 261.

of Quebec. Brooks (*1*) lists the following fruits as recorded hosts: Hawthorn, wild crab, haw, wild cherry, quince, pear, and apple. Petch (*6*) states that it attacks pear, plum, wild crab, cherry, and hawthorn. Watson (*8*) collected it on cotton in Florida. In northern Colorado it has been reported by List [3] as injuring 100 per cent of the sour cherries in certain orchards.

Control measures recommended for the apple curculio have usually been those found effective against the plum curculio. Such advice seems to be based on the similarity of habits of the two insects rather than on experimental data. Riley (*7*) predicts that the apple curculio can never be controlled by the tree-jarring method and says that the only real remedy is the destruction of infested fruit. Gillette (*5*) suggests arsenical sprays for ordinary infestations, supplemented when severe by jarring and the destruction of windfalls by means of hogs or sheep. Brooks (*1*) recommends the destruction of native host plants, jarring, and arsenical sprays, but says that the last will not prove as effective as against the plum curculio. Crandall's (*4*) discussion of control measures is concerned primarily with the plum curculio. Petch (*6*) states that arsenical sprays and dusts will give good results if applied at the right time and that lime sulphur and sulphur act as deterrents. Two popular reports of the control experiments recorded here have been published by R. M. Clark (*2*, *3*) manager of the orchard where the work was conducted.

NATURE OF THE INJURY

The injury to fruit caused by the apple curculio may be locally severe, but it is not universally present in apple orchards, due in part no doubt to the strange reluctance of the insect to feed on apples of many of the common varieties. In many cases the cause of the injury is not known to the orchardist, or is thought to be the work of the plum curculio.

At the time the beetles are feeding very little injury will be noticed on the surface of the fruit. A few small holes like needle punctures may be observed, or the holes may be concealed by small black pellets of excrement. (Fig. 1, *e*, *f*.) If the holes are cut into toward the center of the apple deeper cavities will be found, which are of two types, large oval cavities and slender cavities more or less the shape of the insect's beak. The former are made for the dual purpose of obtaining food and holding an egg; the latter are feeding punctures only and are made largely by the males. (Fig. 1, *g–j*.)

The effect of such punctures is more apparent after considerable growth of the fruit has taken place. The cavity becomes almost completely obliterated within a short time by the rapid growth of the internal tissues of the young apple. This usually results in the destruction of the egg or young grub if the apple remains on the tree. The cavity is closed by the ingrowth of tissue from the sides and bottom, so that a longitudinal section often has the form of an inverted Y. (Fig. 2.) The cavity leaves a lasting imprint upon the growing fruit which becomes evident by the presence of a hardened area extending from the surface toward the core. Growth is inhibited at that point, and as the apple grows a funnel-shaped pit usually develops. (Fig. 1, *o*.)

[3] LIST, G. M. A NEW CHERRY PEST. Rocky Mt. Conf. Ent. Rpt. 4: 5. 1926. [Mimeographed.]

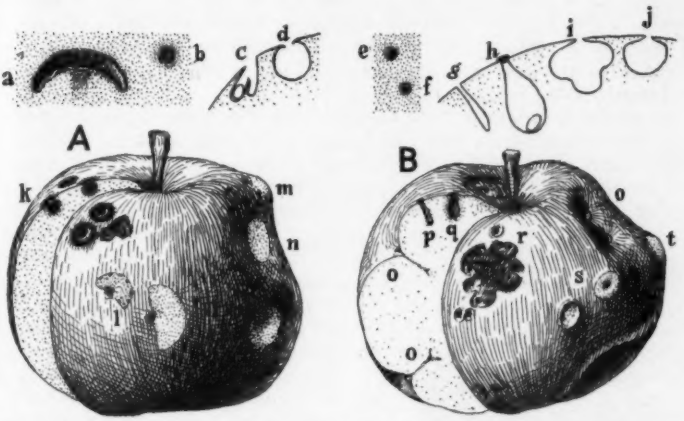

FIG. 1.—A, Injury to apple caused by plum curculio; B, injury to apple caused by apple curculio; *a–j*, recent injury on young fruit, × 3; *k–t*, recent injury on mature fruit and scars resulting from early injury; *a*, crescent cut or egg puncture, surface view; *b*, feeding puncture; *c*, *d*, sections of egg and feeding punctures; *e*, egg puncture plugged with excrement; *f*, same, plug removed; *g*, section of feeding puncture; *h*, section of egg puncture; *i*, *j*, unusual types of punctures; *k*, late summer-feeding punctures, enlarged by drying or rotting; *l*, scars resulting from crescent cuts; *m*, *n*, protruding and sunken scars from early feeding punctures; *o*, pits resulting from egg punctures; *p*, *q*, late summer-feeding punctures; *r*, patch of late summer-feeding punctures resulting in collapse of underlying tissues; *s*, *t*, craterlike and protruding scars resulting from early punctures

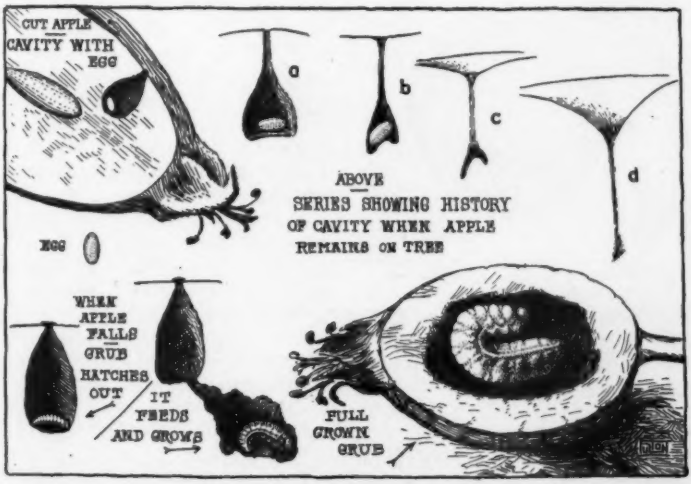

FIG. 2.—Cut apples and sections showing development of apple curculio grub in fallen apples, and history of egg cavities in growing apples

Under some conditions not fully understood the injury may develop as a protruding scar in the form of a rounded bump or a crater. (Fig. 1, *s*, *t*.) Usually an apple which has been affected at all receives a rather large number of punctures, with the result that as it develops it becomes very much deformed and is entirely worthless as a marketable fruit.

An entirely different type of injury results from the feeding of the new generation of beetles which appear in midsummer. It may be found from the middle of July on. The feeding cavities are like those made in the young apples, but they are not closed over by the growth of the apple. Instead, the portion of the skin undermined dries out, enlarging the opening. If the fruit is still quite green the inner part becomes a hardened scar lining the cavity. If the fruit is nearer maturity the pulp surrounding the cavity tends to dry out and shrink, causing the cavity to enlarge further. Usually a number of holes are located close together near the stem or calyx or under a sheltering leaf. In such cases the whole included area of skin dries and turns brown and the material underneath collapses as a result of the drying, so that no distinct cavities are apparent. All that one sees is a sunken and perforated brown patch on the side of the apple. (Fig. 1, *r*, and fig. 3.) The skin may shrivel and crack open, exposing the dried and discolored pulp. This type of injury has been ascribed to plum curculio, but punctures made by this insect in its late feeding are shallower and usually more scattered.

FIG. 3.—Punctures, concentrated under a protecting leaf, made by late-feeding apple curculio

The work of the apple curculio can be distinguished from that of the plum curculio by the greater depth of the cavities formed. In the young fruit while the injuries are still recent the difference is very apparent. The plum curculio's beak is much shorter and broader than that of the apple curculio and the cavity formed is round close to the surface and the hole through the skin is rather large. (Fig. 1, *b*, *d*.) The egg punctures of the plum curculio are very easily recognized on account of the crescent-shaped cut along one side of the egg puncture proper. (Fig. 1, *a*, *c*.)

In the mature apples the plum curculio injuries are more apt to develop as spreading scars and usually lack the hardened internal scar tissue extending toward the core. The feeding punctures become rounded scars and the crescent cuts change to semicircles or half

moons. (Fig. 1, *l*.) These scars often occur in depressions but they are seldom deep funnel-shaped pits. In the case of apple curculio injury the pits rarely have more than a small scar at the bottom.

SEASONAL HISTORY

The apple curculio has but one generation a year and passes most of its life in the adult or beetle stage. It comes out of hibernation in the spring and begins feeding on the young apples just after they have set. Egg laying begins while the apples are still quite small. Most of the eggs are deposited before the June drop is complete, but a few beetles continue laying until the apples are nearly half grown. The egg-laying period covers a month or more. Each female deposits a few eggs a day, averaging 60 to 70 during the season.

Normally the apple must drop if the life history of the insect is to be completed. The entire larval and pupal stages are spent within

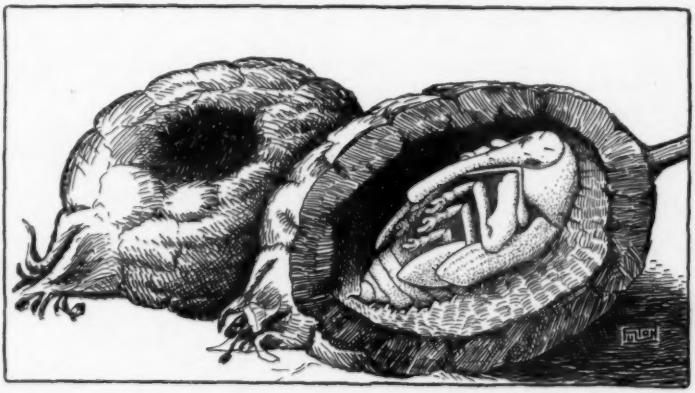

FIG. 4.—Fallen apple from which an adult apple curculio has escaped, and cut apple showing pupa

the dropped apple (fig. 4), which may become dried and mummified before the beetle emerges. Young living larvae were found in some apples picked off the trees in 1926, but none were found in apples picked in 1925. No large larvae have ever been found in growing apples. Evidently the insect is not completely adapted to the cultivated apple and is dependent on the June drop for survival.

A comparison of the percentage of injured fruit on the trees with that on the ground would indicate that the work of the apple curculio has little if any influence on the dropping. Dropped apples collected at several times during 1926 under one group of trees showed an infestation of 37 per cent in a total of 1,073. Counts of injured fruit on the same group of trees taken after the egg-laying season showed 39 per cent injured in a total of 703 apples.

The collecting of beetles and immature stages of the apple curculio has brought out facts concerning the seasonal history which corroborate the results obtained in previous life-history work and which are important in the application of control measures. The adults were found to be present throughout the year but became scarce

early in July because of the dying off of the old generation before many of their offspring had matured. A few of the old generation continued to live and deposited their eggs later in July. In the insectary the last egg was found on July 2 in 1925 and on July 7 in 1926. Eggs were probably deposited in the orchard subsequent to the latter date, for six larvae were collected in fallen apples on August 10, 1926. The transverse diameter of these apples (Ben Davis) was 20, 30, 30, 30, 35, and 38 mm. These larvae were probably from the last eggs deposited that season. Since the apples probably fell soon after the eggs were deposited, their size furnishes an indication of the limits of the egg-laying period. In Crandall's experiments eggs were deposited from May 23 to July 22, a period of over 60 days.

The number of beetles of the new generation leaving the fruit before July 1 must be negligible. The first beetle to be reared to maturity appeared July 3. After the first week in July the beetles began to mature in rather large numbers. By the middle of July 86 per cent had matured from the first dropped apples in 1925 and 100 per cent in 1926. (Table 1.)

TABLE 1.—*Number of dropped and picked apples infested by the apple curculio, and number of curculios in the egg, larva, pupa, and adult stages on different dates in the summers of 1925 and 1926*

Lot No.	Date collected	Date examined	Dropped or picked apples	Apples infested		Curculios in different stages							
						Egg		Larva		Pupa		Adult	
				Number	Per cent	Number	Per cent	Number	Per cent	Number	Per cent	Number	Per cent
1	June 17, 1925	June 18[1]	Dropped			3	19	13	81				
	....do	July 21[1]	....do					42	76	13	24		
	....do	July 14[2]	....do							8	14	51	86
2	....do	June 18[1]	Picked			12	92	1	8				
	....do	July 14[2]	....do					3	17	9	50	6	33
3	June 15, 1926	June 16[1]	Dropped			2	10	18	90				
	....do	June 30[1]	....do	487	33.0			52	32	110	68		
	....do	July 13[2]	....do									1	100
4	July 14, 1926	July 15	Small early drops.	205	39.0			10	15	33	52	21	33
5	....do	....do	Medium-sized drops.	281	46.0			45	36	79	64		
6	....do	....do	Recent drops	100	24.0			24	100				
7	Aug. 9, 1926	Aug. 10	Dropped	205	6.8			7	54	4	31	2	15
8	June 15, 1926	June 16[1]	Picked			26	58	19	42				
	....do	July 1[2]	....do					52	84	10	16		

[1] Part. [2] Remainder.

On account of the extended egg-laying period it is of considerable interest to find out the probable proportion of eggs deposited at different times as the season advances. The heaviest drop of apples comes during a period of a few weeks in June, several weeks after the blossoming period. The proportion of fruit falling during the June drop is quite large except when very few blossoms have set. In 1926 the set of fruit was average or better and the June drop was normal. The apples in lots of 3, 4, 5, 6, and 7 (Table 1) were collected from the same rows of trees and show by the number infested something of the amount of egg laying at the time they fell. Lot 3, which was 33 per cent infested, included the first June drop prior to June

15. Lots 4, 5, and 6 were all collected July 14 and were segregated by size. Lot 4 included the small withered apples which probably fell soon after the middle of June; lot 5 included larger withered apples which probably fell in late June or early July; lot 6 contained only larger fruit which could not have been off the trees much more than a week. The degree of infestation in the three lots was 39 per cent, 46 per cent, and 24 per cent. Lot 7, collected August 9, contained apples which appeared to have fallen so recently that the curculios could not have emerged from them. They were only 6.8 per cent infested.

The percentage of infested apples in lot 6 is still quite high, but the number of apples that dropped in July must have been considerably reduced. The number dropping between the middle of July and August 9 must certainly have been small compared to the June drop. Of the set fruit which falls before the approach of maturity it is probably safe to say that three-fourths drops before the 1st of July in a normal year. Since the degree of infestation is greater before that date it seems probable that 80 per cent of the whole brood of apple curculios are on the ground by July 1. All but a small fraction of the remainder probably appear before the middle of July.

HABITS OF THE ADULTS

On account of the peculiar shape and protective coloration of the beetles, they are extremely difficult to find until the eye has become trained to detect them. They have a habit of posing with beak elevated when anything approaches them (fig. 5), and while standing perfectly motionless they are easily mistaken for a loose bud scale or a cluster of dried petals. If a moving object comes close to them they will drop. The readiness with which they crawl away or take to flight would make a tree-jarring method of control as sometimes used against the plum curculio impractical for this insect.

The adults feed mainly on the fruit, consuming large quantities, especially the females when excavating the egg cavities. To a certain extent the bettles will feed on leaves, making small holes about one-thirty-second of an inch or less. They feed on the cambium of green spurs in much the same way as on the fruit, by enlarging the cavities beneath the surface. This type of feeding was observed in the case of caged insects and its effect can be noted on the orchard trees.

HIBERNATION

An effort was made to find the adult apple curculios in winter to determine where their favorite hibernation quarters are. All search failed of results. Another attempt to solve this problem was made by confining beetles in a large cage under an orchard tree. On August 11, 1926, 80 adult curculios were placed in a cage which contained an old partly decayed apple stump with loose bark, a piece of burlap, dead leaves, matted grass, and a number of apples.

On October 25 the cage was examined and a careful search was made under loose bark, under leaves, and in the upper soil. Fifteen living and two dead curculios were recovered. All were found close to the surface of the ground under matted grass, and most of them were near the door where they had been put in. Apparently they

had not moved about much after August 11. What became of the other 63 beetles is difficult to say, but most of them were probably overlooked in the search. The cage was large, and, as stated before, this insect is difficult to find. None of the apples in the cage had been fed upon.

RESISTANCE OF LARVAE AND PUPAE TO HEAT

One of the control measures usually recommended for the apple curculio is to expose the fallen fruit to sunlight by raking it between the rows or by pruning the trees to let in more sunlight. It was thought advisable to test the effectiveness of sunlight as a killing agent.

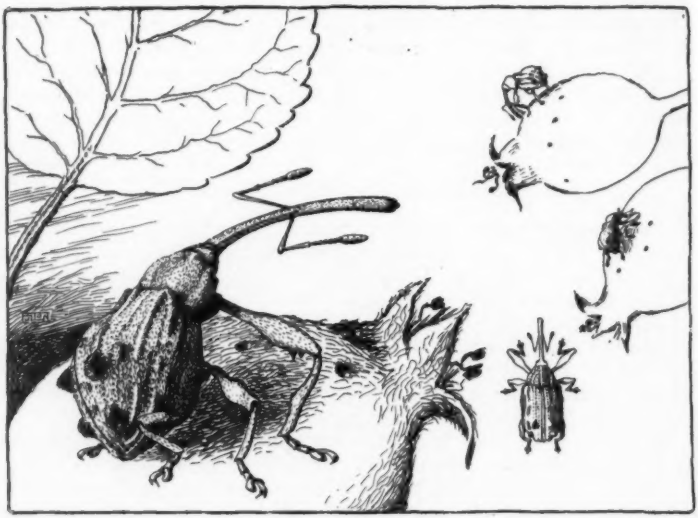

FIG. 5.—Adult apple curculio

On a very hot day a number of infested dropped apples were exposed to the sun by spreading them out in a single layer in a flat pan. The temperature under a tree at the time ranged from 103° to 103½° F. On the surface of the ground in the sun the temperature was 135°; the surface of the pan was 125°–126°, and when the bulb was placed within the cavity of one of the apples the thermometer registered 118° to 120°. The last was probably about the temperature to which the insects were exposed.

After 15 minutes' exposure to the sun, six of the apples were removed and opened. Four larvae were found dead and two alive. After a half hour's exposure another lot was removed and 10 larvae and 5 pupae were found dead. The remaining 9 larvae and 5 purpae were exposed for an hour and all died. As a check, a number of apples from the same lot were examined without exposure to the sun. One dead and apparently parasitized larvae was found among 17 living larvae and 2 pupae.

From this experiment it seems improbable that the insects could live long if the apples were removed from under the trees and exposed to direct sunlight on cultivated ground. Cultivation after the apples have dropped would tend to protect the larvae, however, by shielding them from direct sunlight. Experiments with buried apples showed that the beetles could mature and come out from 2½ inches of loam soil, both loose dry soil and soil that had been water-soaked and dried. Some curculios escaped from 6 inches of soil but ordinary disking probably would not bury apples more than 2 or three inches.

PARASITISM

Parasitism could not have had any marked effect on the degree of infestation of the orchards at Mitchellville. In 1925 only one parasite was discovered in all the material examined. In 1926 the parasites had apparently increased to some extent. In one lot of 235 curculios in various stages, 17 (7.2 per cent) were parasitized. The parasite was a small chalcid fly which was identified by A. B. Gahan as a species of Eurytoma, probably undescribed. The larvae feed externally on the curculio grub. Usually only a withered carcass is found with a full-grown parasite larva. The first adult parasite appeared on July 20, 1926.

CONTROL EXPERIMENTS

Experiments on the control of the apple curculio were all carried on at the Apple Grove Orchards south of Mitchellville, Iowa, or with insects obtained from there. The three orchard plots, separated by intervening field and woods, are described as follows: (1) three acres, 90 per cent Ben Davis variety, woods on three sides; (2) 17 acres, 40 per cent Ben Davis, woods on three sides; (3) main block of 24 acres, 30 per cent Ben Davis, with a spur of 3 additional acres on the southwest corner, 70 per cent Ben Davis. The main block is surrounded by open fields but the spur is directly across the road from a wooded pasture. The apple curculio became severe in orchard No. 2 near the timber about 1915. It spread slowly and by 1925 was most severe in orchard No. 1 and the 3-acre spur of No. 3. The main block of No. 3 remained relatively free but was showing an increase in infestation. The location of the original points of attack would indicate that the curculio came into the orchards from the wood lots, or found more favorable conditions near the woods. The increase in infestation was not due to an overflow of curculios breeding in wild crab apples or hawthorns, however, for such trees had been removed from all woods, except on the north side of orchard No. 1, where the land was under different ownership.

In these orchards the most extensive injury has been done to the Ben Davis variety. A few bearing Delicious in orchard No. 2 have been about as severely injured. Grimes Golden and Willow Twig have been injured to some extent, and Jonathan, Stayman, and Winesap have been slightly injured. Northwestern Greening remained free from injury even when adjacent to the worst infested trees.

Attempts to control the apple curculio by applying an extra lead arsenate spray between the "calyx" and "first-cover" sprays were unsuccessful. The orchards were disked until mid-July, trees were pruned for more sunlight, adjacent fence rows were cleaned up, and

woodland west and north of orchards 3 and 4 were thinned out and pastured. When hogs were available they were turned into the orchard, but this was done only in the spring. These measures resulted in the practically complete elimination of the plum curculio in the orchards but had no lasting effect on the apple curculio. After nine years the latter was more destructive than ever.

In the spring of 1925 an experiment was begun in which hogs were used more intensively. During April and May the orchards were cultivated with a 14-foot extension disk to eliminate as much grass as possible. Pigs weighing from 65 to 90 pounds removed most of the remaining grass under the trees by early June. They then began to feed on the fallen green apples. Until the June drops were well cleaned up in July almost no grain was given them. The number of pigs and the period of pasturage are shown in Table 2 for each orchard.

TABLE 2.—*Number of pigs and period of pasturage in the different orchards in 1925 and 1926*

Orchard No.	Number of acres fenced	Number of pigs pastured in 1925	Period of pasturage in 1925	Number of pigs pastured in 1926	Period of pasturage in 1926
1	3	15	June 25–Aug. 30	0	
2	17	50	June 6–July 20	45	Mar. 1–June 20.
3	3	15	Apr. 1–June 25 and 5 or 6 days in July.		
	24			45	June 20–July 20.

In considering the effect of control measures one must bear in mind that the use of pigs is directed against the new generation of curculios and has practically no effect on the early injury to the fruit. To some extent, then, the amount of injury in the orchard in 1925 could be considered a check on the control obtained for the following year.

The dropped apples were cleaned up very thoroughly in the 3-acre spur of orchard No. 3. Counts of injured and uninjured fruit in this block were made on August 25, 1925. Apples on the lower branches only were counted. On 13 trees there were 1,751 apples. Of these, 1,232 showed injury and 519 showed none; that is, 70 per cent of the fruit was injured. The injury to fruit on individual trees ranged from 40 to 95 per cent.

A very small portion of the injury was caused by the late feeding of the new generation of beetles. This late type of injury had heretofore been quite important in the orchard and added considerably to the total loss.

On July 14, 1926, counts were made in the same part of the orchard. A total of 5,246 apples were examined and of these only 93 (1.77 per cent) showed any injury by apple curculio. The fruit injury of individual trees ranged from 0 to 4.7 per cent.

Orchard No. 1, which had also been thoroughly pastured by hogs in 1925, yielded 500 bushels of apples in 1926, the first paying crop that had been harvested from that block for six years. No counts of injured fruit had been taken in this orchard, but the improvement in the crop was as marked as in No. 3. In 1926 dropped apples containing larvae were scarce, whereas in 1925 an abundance of larvae were obtained by picking up dropped apples at random.

In 1925 orchard No. 2 was only a little less infested than No. 1 and No. 3. A small plot in the southeast corner, separated from the remainder by a gully, was very thoroughly freed from drops in 1925 due to the presence of a wallow in the gully which kept the pigs in that part of the orchard. In 1926 these trees were practically free from curculio injury.

Farther back in the orchard the drops were not so well picked up, and in 1926 the injury in this part was estimated as a check on the figures taken in orchard 3, where the clean-up had been thorough. On August 9, 1926, counts made of injured and uninjured fruit on five trees gave a total of 278 injured and 425 uninjured, or 39 per cent injured. The percentages of injury on the five trees were 16, 28, 32, 43, and 68. Thirty-seven per cent of the dropped apples collected in the same part of the orchard during the season were infested.

The experiments with pasturing pigs were sucessful from a business standpoint. A cost account kept for the two years showed that this method of control was more than economical, for it actually netted a profit. In 1925 each pig returned a net profit of $10 above cost and feed and in 1926 a net profit of $7.65. These figures include the cost of vaccination but not the item of labor in handling or feeding the pigs, nor the cost of fencing, which in his case was small.

A few experiments were tried to determine the possibility of poisoning curculios with arsenical sprays and dusts. Five curculios were caged on a small branch sprayed with calcium arsenate at the strength of 1 pound to 50 gallons of water plus 3 pounds of lime. Five curculios were caged on a second branch sprayed with the same material with the addition of enough molasses to give a slightly brown color. None of the insects showed any sign of poisoning and continued to feed.

On June 5, 1925, 25 curculios were placed in each of three different wire cages over branches of a tree. One branch was thoroughly dusted with calcium arsenate after the cage was in place, so that some of the powder settled on parts of the cage. Another branch was sprayed with the same material at the rate of 1 pound of the poison to 50 gallons of water; the third was untreated. On June 9, the cages were examined but not opened. The one with the dusted branch had 17 dead curculios in the bottom, the one with the sprayed branch had 4 dead, and the check had 1 dead. On June 10 the first two cages were removed. In the cage with the dusted branch 20 curculios were dead and 3 alive, leaving 2 unaccounted for; in the cage with the sprayed branch 4 were dead and 21 alive; in the check cage only 1 was dead. Apples on the check and sprayed branches were riddled with punctures, while those on the dusted branch showed no signs of having been fed upon.

On June 12 the same experiment was repeated in the greenhouse. A small branch was sprayed with calcium arsenate, 1 gm. in 400 c. c. of water, of which about one-fourth was applied. Another branch of similar size was dusted, using 1 gm. of calcium arsenate applied with a small hand duster. Cages were fitted over the branches and 10 curculios were released in each before the sprayed branch was quite dry. On June 15 all curculios were alive and the fruit had many feeding punctures. From the fact that the insects lived so long after the treatment it was apparent that the poison had had little or no effect on them. The success of the dusting in the previous

experiment was possibly due to the large quantity used or to the deposit in the bottom of the cage where the curculios may have crawled about.

As a field experiment, a number of trees were dusted with calcium arsenate, using a rotary fan type of hand duster from a stepladder. On July 2, five days after treatment, the lower branches were shaken over a tarpaulin. The number of curculios collected from the dusted trees were 0, 1, 2, 4, 8, 2, 9, 0, 0, 3. All except one were alive. Only two untreated trees were shaken, yielding a catch of 2 and 1. A few weeks before, 20 or 30 curculios could have been collected easily from one tree. It seemed evident that the small number taken from the treated trees was not due to poisoning, but to natural dying off at this time. The new generation had not yet come out in quantity.

CONCLUSIONS AND RECOMMENDATIONS

The experiments with the use of poisons, supplemented by the observations of Clark in the orchard, show that there is little hope of controlling the apple curculio with arsenical poisons. Crandall (*4*) reached the same conclusion after feeding apple curculios on fruit treated with Paris green.

The experiment with the use of hogs has shown that five pigs per acre can, if properly handled, clean up the early dropped apples in an orchard and thus control the apple curculio. The critical time for such control, as shown by the seasonal history data, is from the middle of June until about the middle of July. Pigs weighing about 100 pounds are the best size for this purpose since they do not tramp down the low branches. They do not feed from the trees to any great extent if the apples are more than a foot above the ground.

The pigs prefer green apples to grass, and they can find the apples more readily if the orchard is cultivated before the middle of June. No cultivating should be done after the apples begin to drop.

Pigs should be encouraged to frequent parts of the orchard containing the varieties most subject to injury by the apple curculio. This can be done by throwing there whatever extra feed is necessary. The best results will be obtained if the pigs are kept on slightly short rations.

The greatest drawback to keeping pigs in the orchard is that they injure the trees by rooting or by rubbing against the trunks. When small pigs are used for only a month such injury is negligible. Pigs should not be oiled while they are in the orchard, on account of possible injury to the trees from oil rubbed into the bark.

It is sometimes difficult to obtain pigs at the time when the pasturing should be done. The fruit grower should therefore anticipate this need and buy them during the winter and spring whenever they can be obtained most economically.

SUMMARY

Both the early and late injuries to apples caused by the apple curculio show certain characteristics which distinguish them from similar injuries by the plum curculio.

Egg punctures formed by the apple curculio are closed up rapidly by the growth of the apple. Larvae survive and mature only in

apples which drop from the trees. There is no evidence to indicate that the presence of eggs or larvae causes dropping.

Probably 80 per cent or more of the total new brood of curculios likely to survive will be found on the ground in the egg, larval, or pupal stage by July 15. Adults of the new generation begin to emerge about the 1st of July.

Curculios in immature stages in the apples are killed by exposure to sunlight on bare ground.

Adults are able to emerge when apples are buried under a few inches of soil.

The apple curculio can not be readily controlled by arsencial sprays, orchard cultivation, or the destruction of wild host plants.

Dropped apples may be effectively destroyed and severe infestations of the apple curculio controlled by confining a sufficient number of hogs in the orchard during early summer.

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THE RELATIVE UTILIZATION OF DIFFERENT CALCIUM COMPOUNDS BY HENS IN THE PRODUCTION OF EGGS [1]

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INTRODUCTION

In another publication [2] the authors presented evidence to show that calcium in rock phosphate, when fed to hens on a grain-bran-tankage ration, is not utilized in the formation of eggshell; at least, not to the same extent as calcium in the form of carbonate. The experiment described herein was planned to determine to what extent the hen can utilize for egg production the calcium in calcium lactate, calcium chloride, calcium sulphate, and pure tricalcium phosphate, as compared with that in calcium carbonate.

EXPERIMENTAL PROCEDURE

On January 1, 1926, 40 one-year-old White Leghorn hens from the same parent stock were placed in four similar houses. These houses were so constructed that large windows faced the south and east, allowing the free entrance of direct sunlight for several hours each day. The hens received a ration consisting of wheat and yellow corn, and skim milk was always at their disposal. Green food, such as cabbage and lettuce leaves, was given twice a week, and the straw litter covering the floor was changed frequently. From January 1 to March 11, 1926, no calcium compound was added to the ration given to the four lots.

A trap-nest record of the eggs was kept throughout the experiment. The eggs laid by each hen each week were weighed together at the end of the week. The shells were then carefully removed and dried for 12 hours at 100° C. in an electric oven, and after cooling they were weighed. The difference between the average weight of the eggs for a given period and the average weight of the shells for the same period is taken as the average weight of the contents.

On March 11, 1926, the hens were weighed individually and divided into 4 lots of 10 each, designated lots 1, 2, 3, and 4. The lots were so divided as to equalize their weights and egg-laying ability. The average weights of the hens in lots 1, 2, 3, and 4 were 1,825, 1,813, 1,826, and 1,825 gm., respectively, and the average egg records for the pullet year were 165, 162, 169, and 164 eggs per hen, respectively.

For the sake of clarity, each lot will be discussed separately. It is to be remembered that all the hens had gone for 70 days without a calcium supplement to their diet, before the calcium compound to be tried was added.

[1] Received for publication Nov. 21, 1927; issued April, 1928. Published with the approval of the director of the Kentucky Agricultural Experiment Station.

[2] BUCKNER, G. D., MARTIN, J. H., and PETER, A. M. CALCIUM METABOLISM IN THE LAYING HEN. Ky. Agr. Expt. Sta. Bul. 250, p. 329-367. 1923.

EXPERIMENTAL DATA

LOT 1

Beginning March 12, 1926, a very high grade limestone (96.3 per cent $CaCO_3$) crushed to about one-fourth inch in size and screened free of dust was kept before the hens constantly until April 11, 1926. During this time the 10 hens consumed 2,680 gm. of the limestone, which is at the rate of 8.7 gm. per hen per day. On April 12 the limestone was replaced by rock gypsum crushed to about the same size as the limestone, screened, and freed from carbonates by treatment with hydrochloric acid and washing free of chlorides. This purified rock gypsum contained 94.6 per cent $CaSO_4.2H_2O$. Between April 12 and July 18, 1926, the hens consumed 6,090 gm. of the purified gypsum, which was at the rate of approximately 6.3 gm. per hen per day, and it was greedily eaten.

TABLE 1.—*Number of eggs produced by hens of lot 1, kept on a grain–skim-milk ration fed without the addition of mineral or with the addition of calcium carbonate or calcium sulphate; together with the average weight of the eggs, shells, and contents*

Additions to basal ration	Number of days of feeding	Number of hens	Total number of eggs	Number of eggs per day per 100 hens	Average weight of 1 egg	Average weight of contents	Average weight of dry eggshell
					Gm.	*Gm.*	*Gm.*
No mineral added:							
Jan. 1–31	31	10.0	18	6	53.3	48.6	4.7
Feb. 1–Mar. 11	39	10.0	41	11	55.9	51.5	4.4
Calcium carbonate added:							
Mar. 12–Apr. 11	31	10.0	179	58	56.9	51.2	5.7
Calcium sulphate substituted for calcium carbonate:							
Apr. 12–30	19	10.0	124	65	57.1	52.3	4.8
May 1–17	17	10.0	102	60	53.4	49.2	4.2
May 18–June 14	28	10.0	104	37	50.5	46.4	4.1
June 15–July 18	34	*9.5	26	8	51.8	47.8	4.0

* 1 hen died July 2.

It will be seen in Table 1 that the average weight of the eggs for the 39 days previous to March 12, 1926, was 55.9 gm., that of the contents, 51.5 gm., and that of the dry shells, 4.4 gm. The addition of calcium carbonate to the ration did not increase the average weight of the contents of the eggs during the following 31 days, but the average weight of the dry eggshells was increased 1.3 gm. After the addition of the carbonate more than five times as many eggs per day were produced. Immediately after the calcium carbonate was replaced by calcium sulphate there was a slight increase in the daily egg production, but after May 18, when production should normally remain high, it diminished rapidly. There was also a lowering in the average weights of the contents and of the dry eggshells. These facts indicate that calcium carbonate is utilized to a much greater extent by the hen for the production of eggs than is calcium sulphate. An examination of the droppings of the hens showed that large quantities of limestone and gypsum were being passed through the hens unchanged while each was being added to the ration.

LOT 2

From March 12 to April 11, 1926, during which time they had free access to the prepared gypsum described under lot 1, the 10 hens consumed 1,300 gm. of that mineral, which is at the rate of 4.2 gm. per hen per day. Soon after the gypsum was added to the diet, the droppings became soft and slightly watery and this condition continued in varying degree. The number of eggs laid by the hens was daily becoming less and the general condition of the birds was not good; so the gypsum was replaced by limestone on April 12. The hens consumed 4,105 gm. of limestone between that date and July 18. This is at the rate of approximately 4.2 gm. of limestone per hen per day.

TABLE 2.—*Number of eggs produced by hens of lot 2, kept on a grain–skim-milk ration fed without the addition of mineral or with the addition of calcium sulphate or calcium carbonate; together with the average weight of the eggs, shells, and contents*

Additions to basal ration	Number of days of feeding	Number of hens	Total number of eggs	Number of eggs per day per 100 hens	Average weight of 1 egg	Average weight of contents	Average weight of dry eggshell
					Gm.	*Gm.*	*Gm.*
No mineral added:							
Jan. 1–31	31	10	13	4	60.9	56.4	4.5
Feb 1–Mar. 11	39	10	48	12	57.8	53.6	4.2
Calcium sulphate added:							
Mar. 12–Apr. 11	31	10	70	23	53.9	49.4	4.5
Calcium carbonate substituted for calcium sulphate:							
Apr. 12–30	19	10	110	58	58.8	53.4	5.4
May 1–17	17	10	108	63	58.2	53.2	5.0
May 18–June 14	28	10	121	43	56.8	51.6	5.2
June 15–July 18	34	9	107	35	57.1	51.9	5.2

In Table 2 it will be seen that the egg production was nearly doubled following the addition of calcium sulphate to the ration of grain and skim milk. However, this increase was considerably less than the increase in egg production caused by the addition of calcium carbonate to the same ration in lot 1. The average weight of contents was 4.2 gm. less for the period the calcium sulphate was added to the diet than for the 39 days preceding, when no calcium supplement was used, whereas the average weight of the dry shells remained practically unchanged. When calcium carbonate was substituted for calcium sulphate, the average weights of the contents and dry shells were increased materially and the number of eggs laid was more than doubled. The decline in egg production following May 18, was slower when calcium carbonate was substituted for calcium sulphate than when the sulphate was substituted for the carbonate in lot 1.

LOT 3

From March 12 to June 14, 1926, the hens received calcium lactate (98.2 per cent) daily in 200 gm. of a wet mash consisting of equal parts of bran, shorts, and corn meal, at the rate of 5 gm. of calcium lactate per hen per day for the first 20 days and 10 gm. per hen per day from then until June 14. When the calcium lactate mash was first given the hens, it was eaten greedily, but on May 18 it was

noticed that the hens were not eating it all and by June 14, it was refused entirely. On June 15 calcium carbonate (limestone) was substituted for the calcium lactate and the hens consumed 740 gm. during the next 34 days, which is at the rate of approximately 2.2 gm. per hen per day.

It is believed that the introduction of the wet mash, as such, into the diets of lots 3 and 4 could not have influenced egg production materially.[3]

TABLE 3.—*Number of eggs produced by hens of lot 3, kept on a grain–skim-milk ration fed without the addition of mineral or with the addition of calcium lactate or calcium carbonate; together with the average weight of the eggs, shells, and contents*

Additions to basal ration	Number of days of feeding	Number of hens	Total number of eggs	Number of eggs per day per 100 hens	Average weight of 1 egg	Average weight of contents	Average weight of dry eggshell
					Gm.	*Gm.*	*Gm.*
No mineral added:							
Jan. 1–31	31	10	16	5	53.9	50.1	3.8
Feb. 1–Mar. 11	39	10	48	12	54.1	49.9	4.2
Calcium lactate added:							
Mar. 12–Apr. 11	31	9	93	33	53.9	49.6	4.3
Apr. 12–30	19	9	89	52	54.6	49.7	4.9
May 1–17	17	9	79	52	54.3	49.6	4.7
May 18–June 14	28	9	81	32	53.4	48.9	4.5
Calcium carbonate substituted for calcium lactate:							
June 15–July 18	34	8	130	48	57.9	52.8	5.1

The addition of calcium lactate to the ration did not materially change the average weight of the contents (Table 3), but caused a slight increase in the average weight of the dry shells. There was, however, a marked increase in the number of eggs laid. When the hens began to refuse to eat the mash on May 18 there was a noticeable fall in egg production. When calcium carbonate was added, egg production was increased and the average weights of the contents and the dry eggshells were greater. It seems therefore that the calcium in calcium lactate can be utilized by the hen for egg production to the extent that it is consumed.

LOT 4

From March 12 to April 5, 1926, 200 gm. of a wet mash consisting of equal parts of bran, shorts, and corn meal, containing 4 gm. of calcium chloride (81.0 per cent) per hen was fed daily. The addition of calcium chloride caused some diarrhea, which was accompanied by loss of appetite for the mash. On April 5 the hens refused to eat any of the mash, and since egg production had not greatly increased, precipitated tricalcium phosphate (99.2 per cent) was substituted for the calcium chloride and was given at the rate of 10 gm. per hen per day in the same wet mash. The mash containing phosphate was eaten greedily and the amount supplied to the hens was consumed completely. Because most of the hens had stopped laying by June 14 limestone (calcium carbonate) was substituted for the tricalcium phosphate. The hens consumed 395 gm. of the limestone, which is at the rate of approximately 1.2 gm. per hen per day.

[3] MARTIN, J. H. SOURCES OF ANIMAL PROTEIN FOR LAYING HENS. Ky. Agr. Expt. Sta. Bul. 260, p. 99–132, illus. 1925.

TABLE 4.—*Number of eggs produced by hens of lot 4, kept on a grain–skim-milk ration fed without the addition of mineral or with the addition of calcium chloride, tricalcium phosphate, or calcium carbonate; together with the average weight of the eggs, shells, and contents*

Additions to basal ration	Number of days of feeding	Number of hens	Total number of eggs	Number of eggs per day per 100 hens	Average weight of 1 egg	Average weight of contents	Average weight of dry eggshell
					Gm.	*Gm.*	*Gm.*
No mineral added:							
Jan. 1–31	31	10	25	8	60.3	55.4	4.9
Feb. 1–Mar. 11	39	10	37	9	56.1	51.6	4.5
Calcium chloride added:							
Mar. 12–Apr. 5	25	9	45	20	55.1	51.0	4.1
Tricalcium phosphate substituted for calcium chloride:							
Apr. 6–30	25	9	59	26	52.9	48.5	4.4
May 1–17	17	8	38	28	53.9	49.1	4.8
May 18–June 14	28	8	51	23	51.9	47.2	4.7
Calcium carbonate substituted for tricalcium phosphate:							
June 15–July 18	34	7	88	37	54.6	49.5	5.1

The results in Table 4 show that the addition of calcium chloride was followed by a small increase in the number of eggs laid and a small lowering in the average weights of the contents and the dry shells. After the substitution of tricalcium phosphate there was some increase in egg production, a decrease in the average weight of the contents, and a slight increase in the average weight of the dry eggshells. When calcium carbonate was substituted for tricalcium phosphate there was an increase in egg production and a noticeable increase in the average weights of the contents and dry shells. This is significant because it occurred at a period (June 15 to July 18) when a decline in egg production and egg weights is to be expected.

In order to bring out more clearly the effects following the addition of the several calcium compounds to the basal ration, Tables 5 and 6 have been prepared. Here are shown the comparative production of eggshell and egg contents during the several feeding periods, taking as unity the average production of all the hens during the 39 days, February 1 to March 11, when they were not receiving added calcium. The figures were obtained by dividing the weight of shell or contents produced per 100 hens per day during a given period by the corresponding weight per 100 hens per day for the 39-day period when no calcium compound was added to the ration. The figures show strikingly the superiority of calcium carbonate to the other calcium compounds used. A temporary increase in production of egg contents following the substitution of calcium sulphate for calcium carbonate in lot 1 is apparent, but the production of shell did not increase correspondingly.

TABLE 5.—*Relative amounts of eggshell produced by hens kept on a grain–skim-milk ration with the addition of different compounds of calcium, the average production of the same hens during the preceding 39 days on the same basal ration without added calcium being taken as unity*

Days of consecutive feeding periods	Lot 1	Lot 2	Lot 3	Lot 4
31	Carbonate, 7.0	Sulphate, 2.2	Lactate, 3.0	Chloride,* 1.7
19	Sulphate, 6.6	Carbonate, 6.6	Lactate, 5.4	Phosphate * 2.4
17	Sulphate, 5.3	Carbonate, 6.6	Lactate, 5.2	Phosphate, 2.8
28	Sulphate, 3.2	Carbonate, 4.7	Lactate, 3.0	Phosphate, 2.3
34	Sulphate, 0.7	Carbonate, 3.8	Carbonate, 5.2	Carbonate, 4.0

* 25 days of feeding.

TABLE 6.—*Relative amounts of egg contents produced by hens kept on a grain–skim-milk ration with the addition of different compounds of calcium, the average production of the same hens during the preceding 39 days on the same basal ration without added calcium being taken as unity*

Days of consecutive feeding periods	Lot 1	Lot 2	Lot 3	Lot 4
31	Carbonate, 5.2	Sulphate, 2.0	Lactate, 2.9	Chloride,* 1.8
19	Sulphate, 6.0	Carbonate, 5.5	Lactate, 4.6	Phosphate,* 2.2
17	Sulphate, 5.2	Carbonate, 5.9	Lactate, 4.5	Phosphate, 2.4
28	Sulphate, 3.0	Carbonate, 3.9	Lactate, 2.8	Phosphate, 1.9
34	Sulphate, 0.7	Carbonate, 3.2	Carbonate, 4.5	Carbonate, 3.2

* 25 days of feeding.

SUMMARY

A study has been made of the comparative effectiveness of calcium carbonate, calcium sulphate, calcium lactate, precipitated tricalcium phosphate, and calcium chloride in the production of eggs when fed to hens as supplements to a wheat, yellow corn, and skim-milk ration.

The results indicate that calcium carbonate is the most effective of the materials tried, judging by the degree to which it was utilized by the hens in the production of eggs, its influence on the weight of the egg contents and shells, and the quantity consumed.

Calcium sulphate is not so effective as calcium carbonate, as shown by a smaller egg production and lower weight of egg contents and shells produced.

The hens seem to utilize calcium lactate readily, as shown by the number of eggs produced. However, the quantity of calcium lactate consumed was variable and small when compared to the consumption of calcium carbonate or calcium sulphate.

Calcium chloride was not consumed by the hens in sufficient quantity to justify definite conclusions. Its addition to the basal ration caused only a very small increase in egg production.

Precipitated tricalcium phosphate does not serve as a source of calcium in the production of eggs to the extent that calcium carbonate does, as shown by the number of eggs produced and by their weight.

ACETIC ACID AS A SOIL DISINFECTANT[1]

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INTRODUCTION

Acetic acid, in vinegar, is one of the most anciently used preservatives. Its toxicity to bacteria and to fungi has been recognized. Wolf and Shunk (*26*)[2] found that it was more toxic to the six species of bacteria investigated by them than was hydrochloric, sulphuric, citric, tartaric, malic, or formic acids. In the experiments of Bitting (*4*) acetic acid was as toxic to fungi as benzoic, boric, or salicylic acids, and it killed *Penicillium expansum*, *Alternaria solani*, and *Oidium lactis* under conditions in which citric, lactic, malic, and tartaric acids only slightly retarded their growth. According to Rideal and Rideal (*20*), acetic acid 0.3 per cent kills *Bacillus typhosus*. Uppal (*24*) found that it was toxic to the conidia of *Phytophthora colocasiae*. Wüthrich (*27*) reported that 0.01 N acetic acid prevented the germination of the conidia of *Phytophthora infestans* and *Plasmopara viticola* and the spores of *Ustilago carbo*. Results secured by Piemeisel (*16*) led him to believe that it is acetic acid which kills the spores of *Ustilago zeae* in silage. Acetic acid, 0.1 per cent, was found by Hitchcock and Carleton (*11*) to prevent the germination of urediniospores of *Puccinia coronata*.

Acetic acid may have a toxic or a stimulatory effect on higher plants. In the experiments of Lövinson (*14*) it retarded the germination of peas. Carr and Havercamp (*6*) found that the weights of plants decreased as the quantity of acetic acid, added to the soil in which they were growing, was increased. Heald (*10*) found that the growth of corn seedlings was prevented by a certain concentration of acetic acid, but that this concentration was greater than the concentration of hydrochloric or sulphuric acids which is toxic to these plants.

In the experiments of Small (*21*) a much larger percentage of cuttings of rose, privet, and veronica rooted in acetic acid, 0.01 per cent, than in water, and root systems became larger in the acid medium. Promsy (*17*, *18*) found that the germination of seeds was hastened and the growth of seedlings increased by the application of acetic acid to the sand in which they were growing.

The toxicity of acetic acid has been considered or explained by True (*23*), Kahlenberg and True (*13*), Heald (*10*), Winslow and Lochridge (*25*), Reichel (*19*), Dunn (*8*), Cohen and Clark (*7*), and Uppal (*24*). Apparently, the toxicity of acetic acid is due partly to the hydrogen ion and largely to the undissociated molecule. Kahlenberg and True (*13*) found that undissociated acetic acid is toxic, and, according to Heald (*10*) and True (*23*), the $C_2H_3O_2$ ions are not

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[2] Reference is made by number (italic) to "Literature cited," p. 279.

toxic. Wolf and Shunk (*26*) found that the limit for growth of *Bacterium tabacum* was P_H 5.9 when the medium was adjusted with acetic acid and P_H 4.6 when the medium was adjusted with malic acid. They concluded that the hydrogen ion alone is not responsible for the toxicity of acetic acid.

Soil may be changed chemically as well as biologically by the application of acetic acid. The rate of solubility of soils was found by Bouyoucos (*5*) to be greater after treatment with acetic acid than after treatment with nitric, hydrochloric, or sulphuric acids. According to Carr and Havercamp (*6*), acetic acid forms soluble and toxic salts with iron and aluminum in the soil, and they considered this to be the cause of the toxicity of acetic acid to plants.

There is, then, evidence that acetic acid is fungicidal and bactericidal, and that it may be either toxic or stimulatory to plants. In these effects it is not unlike formaldehyde.

In this paper, which is a preliminary report, results of experiments are presented which show that acetic acid applied to the soil protected plants against diseases caused by fungi in the soil and at a cost about half that of formaldehyde.

PREVENTION OF BLACK ROOT ROT OF TOBACCO BY ACETIC ACID

The fungus *Thielavia basicola* (Berk. and Br.) Zopf causes severe black root rot of tobacco in some of the less acid soils in the Connecticut Valley. This disease and bed rot or damping off are combated in the tobacco seed beds by treating the soil with steam or with formaldehyde.

Soil known to be infested with *Thielavia basicola* was placed in 2-gallon crocks, and to it was applied 7, 14, and 21 c. c. normal acetic acid per 100 gm. dry weight of soil, with treatments in duplicate. With the acid, enough water was added to bring the soil to 60 per cent of its water-holding capacity, and at that degree of saturation it was maintained.

Tobacco seeds planted in this soil four weeks after the application of the acid germinated well. While the heaviest application of acetic acid interfered somewhat with growth, the beneficial effect of appropriate quantities was apparent. When the average area of leaves of control plants was 0.37 square inch, the average area of leaves of plants in soil treated with acetic acid (7 c. c. normal acid per 100 gm. soil) was 1.86 square inches (planimeter measurements), or five times as large as that of the controls.

When these plants were 7 weeks old, their roots were examined for infection by Thielavia. The results are recorded under the first series in Table 1. Black root rot was severe on control plants, and infection was prevented by acetic acid.

TABLE 1.—*Effect of acetic acid on black root rot of tobacco*

Number of cubic centimeters of N/1 acetic acid applied per 100 gm. of soil	Infection of roots of tobacco by Thielavia	
	First series	Second series
0 (Control)	Severe on all	Severe on all.[a]
7	None on 83 per cent, trace on 17 per cent	None.
14	None	Do.
21	do	Do.

[a] An average of 30 lesions per root.

Tobacco plants, from steamed soil, and therefore free from black root rot, were set in this same soil 12 weeks after the application of the acid. As before, the plants grown in the treated soil grew better than the controls with the exception of those receiving the heaviest application of acetic acid, which was so large as to be somewhat toxic to the plants. Black root rot on control plants seriously interfered with their growth.

Eight weeks after these plants were set, their roots were examined for black root rot; the results are recorded under the second series in Table 1. Infection was severe on all control plants, but there was no infection whatever on plants in soil to which acetic acid had been applied.

Equivalent quantities of citric, tartaric, lactic, and malic acids of the same normality were applied to this soil infested with *Thielavia basicola*, but only acetic acid prevented infection.

It has been found by the writer that orthophosphoric acid applied to soil infested with *Thielavia basicola* is very favorable to infection of tobacco by this fungus. Acetic acid together with orthophosphoric acid was therefore applied to such a soil. The soil was kept watered to 60 per cent of its water-holding capacity, and 18 days after the application of the acids tobacco plants from steamed soil were set in it. Six weeks later their roots were examined for black root rot. Infection was severe on control plants and was very mild on plants in soil to which had been applied 9 c. c. normal acetic acid and 8 c. c. normal orthophosphoric acid per 100 gm. of soil. Infection was much reduced, although not entirely prevented, by acetic acid with orthophosphoric acid, and orthophosphoric acid used alone resulted in infection considerably more severe than on control plants.

In order to secure more information about the relative safety or toxicity of acetic acid to tobacco, acetic acid, 56 per cent, was applied to soil in the field at the rate of 3,000 pounds per acre. Two weeks after the application, tobacco plants were set in this soil. No toxicity to plants resulted, and in the absence of black root rot, plants in soil treated with acetic acid had the same average weight at the end of the season as control plants.

In one series of pot experiments, acetic acid was applied to a well-buffered soil (a water-deposited silt or fine sandy loam) and to a poorly buffered soil (an ice-deposited stony loam). Acetic acid was not toxic to tobacco plants (set one week after the application of the acid) in the well-buffered soil, but was toxic to them in the poorly buffered soil.

EFFECT OF ACETIC ACID ON SOIL REACTION

It has been pointed out by Stephenson (*22*) that such organic acids as acetic acid are too rapidly oxidized in the soil to cause an increase in soil acidity. But since it has been found that acetic acid can prevent black root rot of tobacco, and since Anderson, Osmun, and Doran (*2*) and Morgan and Anderson (*15*) have shown the relation which exists between the P_H value of soil and black root rot of tobacco, it is interesting to see whether or not this effect of acetic acid can be correlated with soil reaction.

Acetic acid was applied to soil having an initial P_H value of 6.1, and the P_H values of this soil were determined[3] thereafter at frequent intervals. The results of these determinations are shown in Table 2.

TABLE 2.—*Effect of acetic acid on the P_H value of soil*

Number of cubic centimeters of N/1 acetic acid applied per 100 gm. of soil	P_H value of soil after—					
	6 days	12 days	15 days	31 days	41 days	63 days
0 (control)	6.1	6.1	6.1	6.1	6.1	6.1
7	4.9	6.1	6.1	6.1	6.1	6.1
14	4.6	5.4	5.9	6.1	6.1	6.1
21	4.4	4.6	4.9	5.0	6.1	6.1

The application of acetic acid temporarily increased the acidity of the soil, but within a few days or a few weeks, depending on the quantity used, the P_H value of the soil to which it was applied reverted to its original value. When normal acetic acid, 7 c. c. per 100 gm. of soil, was applied, the P_H value of the soil was changed from 6.1 to 4.9, but 12 days after the application of the acid the P_H value of this soil was back to 6.1. In the case of the application of normal acetic acid, 14 c. c. per 100 gm. of soil, the P_H value of the soil was changed from 6.1 to 4.6, but 31 days after the application of the acid the P_H value had reverted to 6.1.

When acetic acid, 56 per cent, was applied to soil in the field at the rate of 3,000 pounds per acre, and determinations made 4, 8, and 10 weeks after the application, the P_H value of the soil was found unchanged.

When acetic acid was applied to a well buffered soil and to a poorly buffered soil, the P_H value of neither was changed, as determined 3, 6, and 12 weeks after the application of the acid.

Acetic acid was found to surpass either nitric or sulphuric acid in preventing infection of tobacco by *Thielavia basicola*. But the P_H value of the soil treated with acetic acid was 5.9, which, in the absence of acetic acid, is favorable to this fungus (*2*), while soil to which the inorganic acids were applied became considerably more acid (P_H 5.6 to 5.4). This is in agreement with the results of Cohen and Clark (*7*), who found that the growth of certain organisms was inhibited at P_H 5.45 when acetic acid was used to adjust the reaction of the media, but that these same organisms grew at P_H 5.0 when adjustment was made with hydrochloric acid.

Acetic acid has no lasting effect in increasing soil acidity, and its effect on fungi in the soil is not exerted through the factor of soil reaction.

EFFECT OF ACETIC ACID ON GERMINATION AND DAMPING OFF OF LETTUCE, CUCUMBER, TOMATO, AND TOBACCO

Acetic acid, in the concentrations named in Table 3, or formaldehyde, 1 : 50, was applied to a soil known to be infested with species of Pythium and Rhizoctonia. The soil was 4 inches deep, in flats, and the diluted acetic acid or formaldehyde was applied at the rate of 2 quarts per square foot. The soil was stirred 4 days after appli-

[3] The P_H values of soil were determined colorimetrically with the Stirlen double wedge comparator, using the method described by Anderson and Morgan (*1*).

cation of the chemicals and 10 days after their application seeds were planted. There were planted per flat and for each treatment 70 cucumber seeds, 100 tomato seeds, and 200 lettuce seeds. An equal weight of tobacco seeds was planted in each flat and for each treatment. The soil in all flats was kept at 70 per cent of its water-holding capacity and the flats were in air of high relative humidity.

When it was evident that all seeds had germinated which were going to germinate, the percentage of seeds which failed to germinate in each treatment was determined. The results are recorded in Table 3.

TABLE 3.—*Effect of acetic acid and of formaldehyde on determination and damping off of cucumber, tomato, and lettuce*

Treatment *		Per cent lost								
Pounds of 56 per cent acetic acid in 50 gallons of water	Per cent acetic acid equivalent	By failure to germinate			By damping-off			By failure to germinate and by damping-off.		
		Cucumber	Tomato	Lettuce	Cucumber	Tomato	Lettuce	Cucumber	Tomato	Lettuce
0.00	0.0	50	30	36	25	0	17	75	30	53
2.98	.4	26	13	7	6	0	4	32	13	11
5.95	.8	11	5	7	3	0	1	14	5	8
8.93	1.2	2	3	0	0	0	0	2	3	0
Formaldehyde 1 : 50		2	0	0	0	0	0	2	0	0

* Applied to soil at rate of 2 quarts of the solution per square foot.

In control flats, 50 per cent of the cucumber seed, 30 per cent of the tomato seed, and 36 per cent of the lettuce seed failed to germinate; and the seeds which failed to germinate were found to be attacked by soil fungi. In soil to which 1.2 per cent acetic acid (8.93 pounds of 56 per cent acetic acid in 50 gallons of water) was applied, none of the lettuce seed, only 2 per cent of the cucumber seed, and 3 per cent of the tomato seed failed to germinate.

There was considerable loss of seedlings of cucumber, lettuce, and tobacco (but not tomato) by damping off in untreated soil. Species of Pythium and Rhizoctonia were present in the damped off seedlings. In the untreated soil 25 per cent of the cucumber seedlings and 17 per cent of the lettuce seedlings damped off. There was no damping off of cucumber and lettuce seedlings in soil to which acetic acid, 1.2 per cent, was applied. The application of this strength of acetic acid was as efficient as formaldehyde in preventing damping off. In the untreated soil 75 per cent of the cucumbers, 30 per cent of the tomatoes, and 53 per cent of the lettuce plants were lost by failure to germinate and by damping off combined. In soil to which 1.2 per cent acetic acid was applied, the loss from these causes was only 2 per cent of the cucumbers, 3 per cent of the tomatoes, and none of the lettuce. Acetic acid 0.8 per cent was nearly as effective in preventing damping off.

In Figure 1 the effect of acetic acid in preventing damping off of tobacco seedlings is evident. Acetic acid 0.4 per cent was insufficient for protection but acetic acid 0.8 or 1.2 per cent prevented damping off.

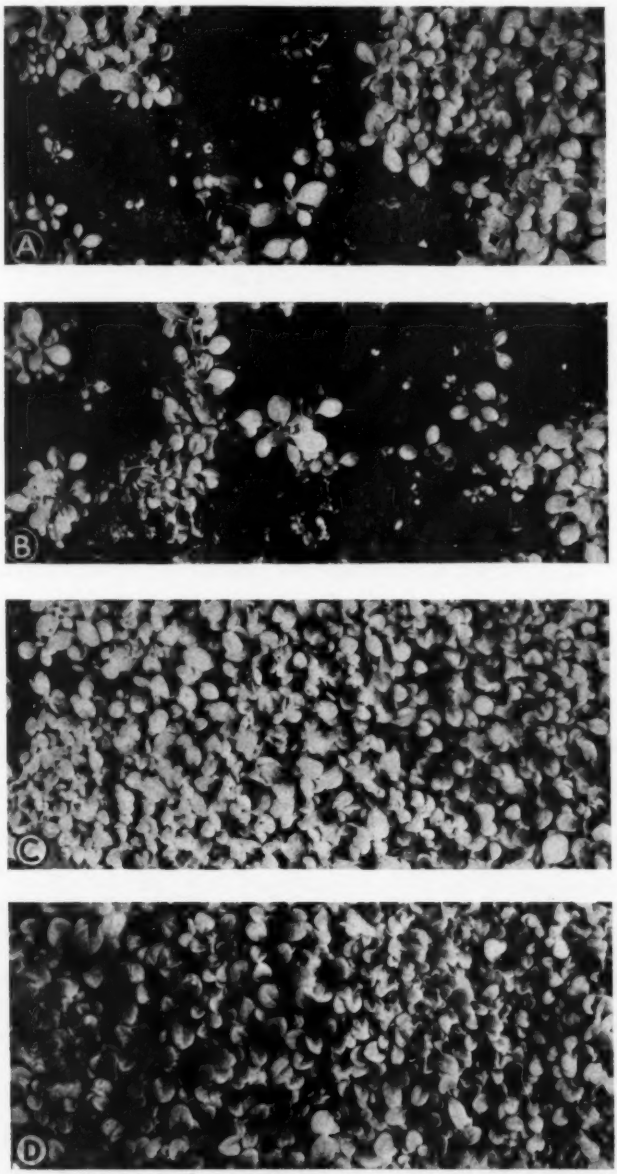

FIG. 1.—Control of damping off of tobacco by the use of acetic acid. A, No acetic acid; B, acetic acid, 0.4 per cent; C, acetic acid, 0.8 per cent; D, acetic acid, 1.2 per cent

Tobacco plants which did not damp off in each of the several treatments were counted. For each 100 plants living in the untreated soil, there were 367 plants living in the soil treated with acetic acid 0.8 per cent and slightly more than this in the soil treated with acetic acid 1.2 per cent, which treatment gave results equal to those secured with formaldehyde.

EFFECT OF ACETIC ACID ON BED ROT AND ON BLACK ROOT ROT OF TOBACCO IN SEED BEDS

Acetic acid, at the rate of 2 quarts per square foot of a solution of 7.5 pounds of 56 per cent acetic acid in 50 gallons of water (equivalent to acetic acid 1.008 per cent), was applied to tobacco seed beds known to be infested with *Thielavia basicola* and with bed-rot fungi, *Rhizoctonia solani* and Pythium sp.[4]

TABLE 4.—*Effect of acetic acid on black root rot and on bed rot of tobacco in seed beds*

Seed bed No.	Per cent of plants showing black root rot		Number of centers of infection of bed rot	
	In control	In acetic-acid treated area	In control	In acetic-acid treated area
1	100	19	0	0
2	26	0	15	0
3	18	0	0	0

In seed beds No. 1 and No. 2 (see Table 4) tobacco seeds were sowed one week after the application of the acid, and in these plots germination was not as good as in the control plots. In bed No. 3, seed was sowed 20 days after the application of the acid, and germination was better in this plot than in its control plot. On the basis of this and other experiments it seems unsafe to sow tobacco seeds as soon as one week after the application of acetic acid to soil. Many weeds came up in the control plots in these seed beds, but very few weeds came up in the plots to which acetic acid was applied.

Bed rot, caused by *Rhizoctonia solani* and Pythium sp., developed in only one seed bed (No. 2). In the control plot of this seed bed, 15 centers of infection appeared and from these bed rot spread rapidly within the plot. There was no bed rot in the plot to which acetic acid was applied. In this and the other seed beds, control plots were of the same area as treated plots and adjacent to them.

The percentages of plants found to be infected with black root rot are recorded in Table 4. In the control plots of seed beds No. 2 and No. 3, 26 and 18 per cent of the plants, respectively, were infected by *Thielavia basicola*. There was no infection in the plots to which acetic acid was applied in these beds. In seed bed No. 1, infection was unusually severe in the control plot, for in it 100 per cent of the plants had black root rot. When acetic acid was applied in seed bed No. 1, there was already so much water in the soil that much—probably one-third—of the solution of acetic acid ran off the bed. Under these conditions black root rot was not entirely prevented by

[4] Acknowledgment is made of the cooperation of Walter W. Sanderson and T. L. Warner, tobacco growers, on whose farm these experiments were conducted.

acetic acid but was reduced to 19 per cent as compared with 100 per cent in the control plot.

In order to learn whether or not dilute acetic acid can be applied to living tobacco plants in the seed bed to control bed rot, acetic acid in several concentrations was sprayed on growing tobacco seedlings among which bed rot had appeared. The spread of the disease was checked, but living plants were injured by the application of a 0.5 per cent solution of the acid. Concentrations so low as not to injure the plants did not prevent the spread of bed rot. The usefulness of acetic acid, as of formaldehyde, for the control of bed rot of tobacco in seed beds is confined to its application to the soil before plants are growing in it.

EFFECT OF ACETIC ACID ON DAMPING OFF OF SEEDLINGS OF WHITE SPRUCE

Acetic acid of two concentrations, 8.33 pounds and 12.5 pounds, of 56 per cent acetic acid in 50 gallons of water (equivalent respectively to 1.12 and 1.68 per cent acetic acid) was applied at the rate of 1.64 quarts per square foot to seed beds in a forest nursery.[5] A control seed bed, not treated, was adjacent to them. Seed beds were 44 square feet in area. One-half pound of seeds of white spruce was planted in each bed seven days after treatment.

Although the interval of time between the application of acetic acid and the planting of seeds was relatively short, germination was not injured. Germination of white spruce seeds was much better and there were fewer weeds in the beds to which acetic acid was applied than in the control bed. Loss by damping off was severe in the control bed, as may be seen by reference to Figure 2, A. There was very little damping off in the beds to which acetic acid was applied. Four months after seeds were planted, the number of seedlings in 5 representative square feet in each bed were counted. The average number of seedlings per square foot in each bed and the number of seedlings in each seed bed figured from this are recorded in Table 5.

TABLE 5.—*Effect of acetic acid on damping off of white spruce seedlings*

Treatment [a]		Average number of seedlings		Seedlings per unit area, expressed in relative numbers
Pounds of 56 per cent acetic acid in 50 gallons of water	Equivalent per cent of acetic acid	Per square foot	Per seed bed	
8.33	1.12	746	32,824	315
12.50	1.68	665	29,260	281
0 (control)	0	237	10,428	100

[a] 1.64 quarts of diluted acetic acid per square foot.

For each 100 seedlings in the untreated soil there were 315 seedlings in soil to which 1.12 per cent acetic acid was applied. This concentration of acetic acid gave better results than the higher concentration (1.68 per cent acetic acid) for in soil to which the higher con-

[5] The cooperation of John Palmer, superintendent of the State forest nursery in Amherst, Mass., is acknowledged.

centration was applied there were 281 seedlings for each 100 seedlings in untreated soil.

The stands in the several seed beds three months after seeds were sowed may be compared by reference to Figure 2. The control of damping off secured by both concentrations of acetic acid is evident, and it is likewise evident that the optimum concentration of acetic

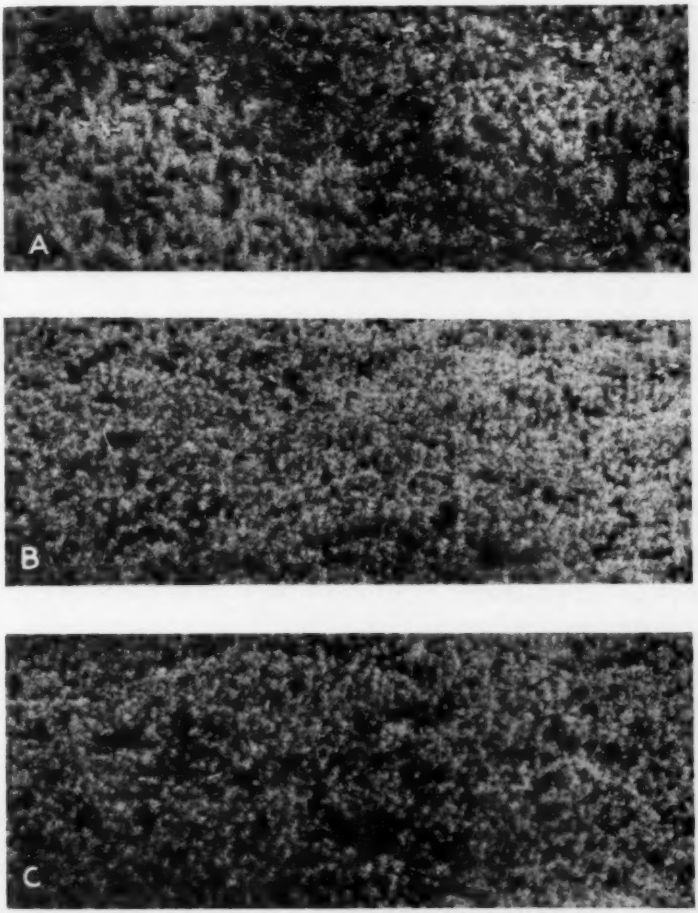

FIG. 2.—Control of damping off of white spruce seedlings by the use of acetic acid. A, No acetic acid; B, acetic acid, 1.12 per cent; C, acetic acid, 1.68 per cent

acid is nearer 1.12 per cent than 1.68 per cent. The fact that acetic acid 1.12 per cent used in this way protected seedlings of white spruce against damping off without injuring the seeds or seedlings is of interest because of the results secured by Hansen, Kenety, Wiggin, and Stakman (*9*). In their experiments, the application of sulphuric acid, hydrochloric acid, or formaldehyde greatly reduced the ger-

mination of seeds of white spruce, and they considered as questionable the use of any of the common fungicides for the prevention of damping off of seedlings of this species.

EFFECT OF ACETIC ACID ON BROWN ROOT ROT OF TOBACCO

When acetic acid was applied to "brown root-rot soil," that is, soil known to have in it the cause of brown root rot, tobacco plants subsequently set in the treated soil grew without showing symptoms of brown root rot. Johnson, Slagg, and Murwin (*12*) found that when formaldehyde was similarly applied to brown root-rot soil, the ability of this soil to produce symptoms of brown root rot of tobacco was destroyed.

The cause of brown root rot is at present unknown, and the results with acetic acid are presented here not because of any light they may shed on the nature of the disease, but because there is in them additional evidence of the effectiveness of acetic acid as a substitute for formaldehyde for soil treatment.

Tobacco seeds were sowed in pots of brown root-rot soil three weeks after the application to it of 1 per cent acetic acid. Seeds were also sowed in pots of this soil without acetic acid. Brown root rot was found on seedlings in untreated soil when they were 4 weeks old. Roots of all plants were examined when they were 9 weeks old. Brown root rot was found to be severe on all plants in untreated soil, but there was not even a trace of the disease on plants in soil to which acetic acid had been applied.

The severity of brown root rot on plants in untreated soil seriously retarded their growth. When the average dry weight of plants in untreated soil was 0.90 gm., the average dry weight of plants in soil treated with acetic acid was 4.76 gm., or more than five times as great as that of the controls.

COST OF USING ACETIC ACID COMPARED WITH COST OF USING FORMALDEHYDE

A cheaper chemical than formaldehyde for the partial sterilization of soil is desirable. Beach (*3*) applied formaldehyde at the usual rate and reported that it cost (in 1926) $0.21 for the 1.4 pounds used to treat 18 square feet of soil, or $508 per acre.

One-half gallon of dilute acetic acid per square foot of soil has been found to be sufficient, and this is the rate at which formaldehyde 1 : 50 is usually applied. The relative cost of these chemicals may therefore be compared by comparing the cost of 50 gallons of each.

Formaldehyde 1 : 50 contains 1 gallon (9.1 pounds) of the commercial 40 per cent solution of formaldehyde in 50 gallons of water. This is now purchasable (in lots of 100 pounds) for about $0.12 per pound. The cost of formaldehyde for 50 gallons (enough for 100 square feet) is therefore $1.09 and the cost of formaldehyde for 1 acre is $475.

The concentrations of acetic acid which were found to control the plant diseases named in this paper were 1.2 to 1 per cent. These are equivalent to 8.93 to 7.44 pounds of 56 per cent acetic acid in 50 gallons of water. Acetic acid of several different concentrations (28 to 99 per cent) is sold, but the cost per unit of actual acetic acid is not greatly different. Acetic acid 56 per cent is now sold (in lots

of 100 pounds) for $0.066 per pound. When 8 pounds of this chemical is used in 50 gallons of water (equivalent to 1.07 per cent acetic acid) the cost of acetic acid for 50 gallons (enough for 100 square feet) is $0.53 and the cost of acetic acid for 1 acre is $231.

The cost of acetic acid is therefore about 49 per cent of the cost of formaldehyde for soil treatment.

SUMMARY

The application to soil of 1 to 1.2 per cent acetic acid (equivalent to 7.44 to 8.93 pounds of 56 per cent acetic acid in 50 gallons of water) at the rate of about one-half gallon per square foot was found to protect tobacco against black root rot, brown root rot, and bed rot or damping off; and to protect cucumber, tomato, lettuce, and white spruce against injury by damping off during and after seed germination.

For the treatment of a unit area of soil, the cost of acetic acid is about 49 per cent of the cost of formaldehyde.

Acetic acid is toxic to seeds and plants with which it comes in contact. The exact time interval which must elapse between the application of the chemical to soil and the planting of seed will depend upon the species of plant and upon the soil; 14, 10, or, in some cases, 7 days have been long enough.

Acetic acid has no lasting effect on the reaction, i. e., the P_H value of soil to which it is applied.

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THE GROWTH OF TOBACCO AND BROWN ROOT ROT OF TOBACCO AS AFFECTED BY TIMOTHY INFUSIONS OF DIFFERENT AGES[1]

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INTRODUCTION

When tobacco is grown after timothy or certain other plants, used either as a cover crop or in the rotation, the growth of the tobacco is often retarded, and a certain accompanying type of injury to the roots of the tobacco has given rise to the name "brown root rot." This apparent injurious effect on tobacco of the decomposing residue of timothy in the soil has frequently been observed and recorded in the literature (*1, 2, 7, 11, 12*).[2]

But this injurious effect of timothy on the tobacco which follows it has been found to be usually transitory by those who have investigated it in field experiments (*1, 2, 7, 11*); that is, the injurious factor which is associated with timothy soon becomes inactive. When the use of a timothy cover crop was discontinued in the experiments of Anderson, Osmun, and Doran (*2*), its previously harmful effect on tobacco quickly disappeared and the yield of tobacco on those plots which had had a timothy cover crop became greater by 17 per cent than on those which had not. Fall plowing of grass sod is reported by Anderson (*1*) to have reduced the injurious effect of sod on tobacco, and in his experiments there was, on plots not fertilized, a difference in favor of plowing under cover crops very early in the spring instead of six weeks later.

The direct cause of brown root rot is at present not known. In the investigations of Johnson, Slagg, and Murwin (*11*), no causal organism was demonstrated to be associated with it.

One hypothesis is that brown root rot is the expression of the toxic effect on roots of tobacco of the decomposition products of certain preceding crops. If such is the case it is to be expected that the effect on tobacco plants of the residues of these preceding crops will depend on the stage of their decomposition. As Martin (*13*) has pointed out, the breakdown of vegetable organic matter includes several distinct steps and consequent intermediary products, and Gardner (*6*) concluded that toxins which result from and during the decomposition of plant residues are, in subsequent stages of the decomposition process, rendered harmless.

Experiments here described were undertaken with the object of learning whether or not there is a relation, such as field experiments have indicated may exist, between brown root rot of tobacco and the stage of decomposition of timothy.

This paper is a record of progress and the results reported should not be considered conclusive.

[1] Received for publication Jan. 9, 1928; issued April, 1928. Published with the approval of the director, Massachusetts Agricultural Experiment Station.

[2] Reference is made by number (italic) to "Literature cited," p. 287.

METHOD

Infusions of timothy plants were made at intervals of one week. Infusions differed only in age and each consisted of the timothy from 2 square feet of sod, washed free of soil and placed in 10 liters of tap water in open jars. Entire timothy plants were used in the first experiment. In the second experiment the tops of the timothy which was used had been kept clipped during its growth. In the first experiment the infusions were stored at about 27° C. and in the second experiment they were stored at about 16° C. To insure the absence of brown root rot, except such as might be due to the application of the infusions, soil was steamed (*11*). Seven kilograms of soil was placed in each of the 2-gallon pots used and five young tobacco seedlings from steamed soil were set in each pot. Treatments were in duplicate so that for each treatment there were 10 plants. No fertilizer was used. The soil in all pots was kept at 60 per cent of its water-holding capacity.

To the soil in each pot, 1 liter of timothy infusion of the ages shown in Tables 1 and 2 was applied when plants were set and 250 c. c. of an infusion of the same age was applied to each pot each week thereafter. Pot No. 1 always received an infusion 1 week old, pot No. 2 an infusion 2 weeks old, and so on. A portion of the infusion which was 1 week old at the first application was applied the next week to other plants as an infusion 2 weeks old, and so on, through the series. All the pots of plants therefore received some of the infusion, but in each case the infusion was of a different age.

EFFECT OF TIMOTHY INFUSIONS ON TOBACCO IN THE FIRST EXPERIMENT

As early as three weeks after plants were set, it was apparent that those in soil receiving infusions 5 to 8 weeks old, inclusive, were making a much poorer growth than plants which were receiving timothy infusions 1, 2, or 3 weeks old.

The writer has several times observed that the symptoms of brown root rot on tobacco seedlings even in "brown root-rot soil" may appear as a retardation of growth several days or even two weeks before the brown discolorations and lesions appear on the roots. This in itself may have some significance as an indication of the nature of the cause of brown root rot when considered in connection with certain other evidence presented in this paper.

Because of the delay which there may be in the appearance of the symptoms on the roots, plants were not removed from the soil until they had been receiving applications of infusions for nine weeks. At that time, they were washed, the roots examined, and the plants dried to constant weight and weighed. The average dry weight per plant in each treatment is recorded in Table 1.

TABLE 1.—*Effect of timothy infusions of different ages on dry weights of tobacco plants in the first experiment*

Age in weeks of timothy infusion applied	Average dry weight (grams) per plant [a]		Dry weights expressd in relative numbers	
	In each treatment	Treatments grouped	In each treatment	Treatments grouped
0 [b]	4.23	4.23	100	100
1	3.96		94	
2	4.53	4.41	107	104
3	4.76		112	
4	3.08	3.08	73	73
5	.45	.91	11	20
6	1.38		30	
7	.08	.08	2	2
8	1.24	1.24	29	29
9	2.58	2.92	61	69
10	3.27		77	

[a] Average of 10 plants per treatment. [b] Water only.

The outstanding result was that the effect of infusions of timothy on the growth of tobacco was found to depend entirely on the age of the infusion. In the comparisons shown in Table 1 dry weights are expressed as relative numbers. Plants treated with infusions 1 to 3 weeks old, inclusive, had an average relative weight of 104 as compared with 100 for plants which received only water. It is therefore apparent that infusions 1 to 3 weeks old did not interfere with the growth of tobacco. Plants to which infusions 4 weeks old were applied had an average relative weight of 73. The application of infusions 5 and 6 weeks old resulted in much smaller plants, with an average relative weight of 20. The smallest plants, which had an average relative weight of only 2, were those to which timothy infusions 7 weeks old were applied. The infusion 8 weeks old was less injurious and plants to which it was applied had an average relative weight of 29. Plants which received infusions 9 and 10 weeks old were significantly larger, with an average relative weight of 69.

Roots on plants which received only water or timothy infusions 1, 2, and 3 weeks old were white and apparently normal.

The roots of all plants in the other treatments were more or less discolored and showed distinct or coalescing brown lesions. In the absence of a proven parasite, the identification of brown root rot depends on symptoms, macroscopic or microscopic. The injury to roots of tobacco plants to which infusions of timothy of certain ages were applied did not differ from the symptoms of brown root rot as it has been described (*11*) and as the writer recognizes it in the field.

There was only a trace of such injury on roots of plants which received infusions 4 weeks old. It was most severe on roots of plants which received infusions 5 to 8 weeks old, inclusive, and was relatively mild on roots of plants which received infusions 9 and 10 weeks old. As in the case of growth, root injury or freedom from it depended on the age of the timothy infusions applied, and the trends of growth depression and root injury were practically parallel.

Other investigators have observed the discoloration or browning of roots of other plants which resulted from similar treatments. Roots

of plants to which Collison (*4*) applied water extracts of straw usually became brown, and Hill (*9*) observed that the roots of plants which received products of the decomposition of cellulose became discolored.

EFFECT OF TIMOTHY INFUSIONS ON TOBACCO IN THE SECOND EXPERIMENT

When the plants in the second experiment had been growing 4 weeks it was apparent that plants which were receiving infusions 2 to 6 weeks old, inclusive, were making a very much better growth and that those plants which were receiving infusions 9 to 12 weeks old, inclusive, were making a poorer growth than plants receiving only water. Two plants were removed from each pot at this time. There were some brown lesions on the roots of the plants the growth of which was retarded, but the injury was not as pronounced as when the remaining plants were examined four weeks later.

The average dry weights of the tobacco plants after timothy infusions had been applied to them eight weeks are recorded in Table 2.

TABLE 2.—*Effect of timothy infusions of different ages on dry weights of tobacco plants in the second experiment*

Age in weeks of timothy infusion applied	Average dry weight (grams) per plant [a]		Dry weights expressed in relative numbers	
	In each treatment	Treatments grouped	In each treatment	Treatments grouped
0 [b]	0.11	0.11	100	100
1	.05	.05	45	45
2	3.92		3,563	
3	3.46		3,145	
4	5.57	4.46	5,063	4,054
5	5.06		4,600	
6	4.29		3,900	
7	.66	.66	600	600
8	.42	.42	381	381
9	.09	.09	82	82
10	.05		45	
11	.05	.05	45	48
12	.06		54	

[a] Average of 6 plants in each treatment. [b] Water only.

The plants did not make a good growth in pots to which only water was applied. The soil was rather poor and no fertilizer was used. But, as may be seen by reference to Table 2, the nature of the effect of the timothy infusions on the growth of tobacco depended, as in the first experiment, on the age of the infusions and hence the stage of the decomposition of the timothy.

However, reference to Tables 1 and 2 will show that a timothy infusion of a certain age did not have the same effect on plants in both experiments. This is not surprising, for the infusions were prepared differently. The proportion of roots to tops of timothy used was greater in the second than in the first experiment, and Garner, Lunn, and Brown (*7*) have found that the roots and tops of preceding plants do not have the same effect on tobacco. Furthermore, the infusions used in the first experiment were stored at a temperature higher by 11° C. that that in the second experiment,

and it is well known that bacterial action similar to that of which such a decomposition is the result, is much affected by temperature.

As shown in Table 2, comparisons of dry weights are expressed as relative numbers, 100 being taken as the average relative weight of plants which received only water. On this basis the average relative weight of plants to which a timothy infusion 1 week old was applied was 45. Plants were greatly benefited by the application of infusions 2 to 6 weeks old, inclusive, so that their average relative weight was 4,054. There was a slight but greatly diminished beneficial effect in the case of plants which received timothy infusions 7 and 8 weeks old, for their average relative weights were 600 and 381, respectively. Infusions 8 weeks old retarded growth considerably and resulted in an average relative weight of 82. Infusions 10 to 12 weeks old, inclusive, were very injurious and the plants to which they were applied had an average relative weight of only 48.

Roots of plants to which only water was applied were white although small. Roots of plants which received timothy infusion 2 to 6 weeks old, inclusive, were white and showed no trace of injury. There were brown lesions and injury, apparently the same as in the first experiment, on roots of plants which received infusions 1 week and 7 to 12 weeks old, inclusive. This injury was most severe on roots of plants which received infusions 1, 9, 10, 11, and 12 weeks old.

DISCUSSION

When infusions of timothy were prepared as in the first experiment, the products of the decomposition of timothy during the first three weeks of the decomposition process were harmless or slightly beneficial to tobacco. These decomposition products were increasingly injurious to tobacco from the fourth to the seventh week, and decreasingly injurious from the seventh to the tenth week of the decomposition. When infusions were prepared by the method used in the second experiment, the products of the decomposition of timothy were injurious the first week, very beneficial from the third to the sixth week, decreasingly beneficial in the seventh and eighth weeks, and decidedly injurious to tobacco in the tenth, eleventh, and twelfth week of the decomposition process. In both experiments the products of the decomposition of timothy were at times innocuous and at times harmful to tobacco, depending on the age of the infusions.

The relation of the response of plants to the stage of the decomposition of vegetable organic matter applied to them has been noted by other investigators. Breazeale (*3*) found that the nature of the effect on plants of the decomposition products of sorghum depends on how far the decomposition process has progressed. In his experiments Collison (*4*) found that the effect produced by extracts of straw on seedlings when fresh extracts were used was different from the effect produced when he used old extracts. Hartwell and Pember (*8*) found that in the early stages of the decomposition of oats, the effect on other plants was unfavorable, but that in later stages of the decomposition there was a favorable effect. Gardner (*6*) concluded that some organic compounds which when fresh and unchanged may be actually toxic to plants are, in most soils, eventually decomposed or otherwise rendered nontoxic, usually by organisms.

But Gardner concedes that some toxins may be decomposed by chemical agencies and this point also is of interest, for formalin has been found (*11*) to destroy the ability of "brown root-rot soil" to produce the disease, and the writer has found that soil treatment with acetic acid has a similar effect.

The fact that only at certain stages in its decomposition did timothy retard the growth of tobacco and apparently induce brown root rot seems to the writer to lend support to the hypothesis that brown root rot may be only the expression on tobacco roots of the injurious effect of the preceding crop. It has been found (*11*) that merely drying or aerating "brown root-rot soil" frees it of the cause of the disease, and this, too, is in agreement with the hypothesis that brown root rot is caused by some toxic substance which is easily volatilized, decomposed, or otherwise rendered innocuous. Several previous investigators (*4*, *5*, *9*) have found that decomposing plant residues contain substances injurious to the roots of plants, and Collison (*4*) concluded that the residues of timothy and certain other plants may be important contributors to soils of compounds toxic to plant growth.

Timothy was used in these experiments because it is one of the most injurious crops in its effect on tobacco in the field. Collison (*4*) observed that extracts of timothy were more injurious to the plants used by him than were the extracts of wheat or oats. There is a possibility that this may be explained by the relative slowness with which timothy decomposes and the consequently long persistence of toxic substances associated with its decomposition. Nonlegumes in general have been found (*9*) to decompose more slowly than legumes, and the decomposition of timothy residues has been found (*14*) to extend over a longer period than that of clover residues.

In the experiments here described infusions of certain ages were found to be beneficial rather than harmful to tobacco. It was one of the conclusions of Jensen (*10*) and of Martin (*13*) that some of the products of the decay of plant substance may render available to plants certain otherwise insoluble nutrients in the soil. The response of tobacco plants in these experiments showed that at some stage or stages in the decomposition of timothy there are products formed which are favorable to tobacco and which certainly do not induce brown root rot.

SUMMARY

When infusions of timothy were applied to soil in which tobacco plants were growing, the effect on the tobacco was found to be sometimes harmless and sometimes very injurious, depending on the age of the infusions; that is, on the stage of the decomposition of the timothy.

The response of plants to an infusion of a given age was found to be influenced by the proportion of tops to roots of timothy used or by the temperature at which the decomposition process went on.

Those infusions of timothy which retarded the growth of tobacco plants produced on their roots brown discolorations and lesions apparently of the same type as those which, in the field, have given rise to the name "brown root rot."

These results lend support to the hypothesis that brown root rot of tobacco is the expression of the injurious effect on tobacco roots of one or more toxic substances which are formed from, and at certain stages in, the decomposition of vegetable organic matter, more especially the residues of certain slowly decomposing crops such as timothy.

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